



WORLD
PRECISION
INSTRUMENTS

INSTRUCTION MANUAL

REMS AutoSampler

Automated TEER Measurement

(version 6.02)

Serial No. _____

www.wpiinc.com

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ABOUT THIS MANUAL

The following symbols are used in this guide:



This symbol indicates a CAUTION. Cautions warn against actions that can cause damage to equipment. Please read these carefully.



This symbol indicates a WARNING. Warnings alert you to actions that can cause personal injury or pose a physical threat. Please read these carefully.

NOTES and **TIPS** contain helpful information.

INTRODUCTION

The **REMS AutoSampler** automates measurements of electrical resistance of transepithelial, transendothelial or Caco-2 cell membranes being grown to confluence on microporous filters of high throughput screening (HTS) 24- and 96-well microplates. It is a PC-controlled, tissue resistance measurement system that offers reproducibility, accuracy and flexibility for this kind of measurement. Automated measurement of tissue resistance in cell culture microplates provides the important advantages of speed, precision, decreased opportunity for contamination and the instant availability of measured resistance data on a computer. These measurements are useful in applications such as drug bioavailability studies and studies on the mechanisms of drug transport.

The **REMS AutoSampler** automates TEER measurements previously made with WPI's **EVOM²** Epithelial Voltohmmeter. Automated tissue resistance measurements up to 20k Ω can be performed on 24- or 96-well HTS microplates. Currently, the REMS is compatible with Corning Costar HTS Transwell-96- or 24 well plates, Millipore Multiscreen™ CaCo 96-well plates and BD Falcon 96- or 24-well plates.

The **REMS AutoSampler** automatically measures and records tissue resistance from a user-specified matrix of culture wells on the microplate. According to the specified sequence, the robotic arm moves into the identified wells taking TEER measurements. By means of a X-Y-Z locating system, the electrode arm is positioned precisely and reproducibly over each well.



Fig. 1 — The REMS AutoSampler includes the computer with all the hardware and software installed, the REMS Interface Unit, the robotic arm, a dummy electrode and one measurement electrode.

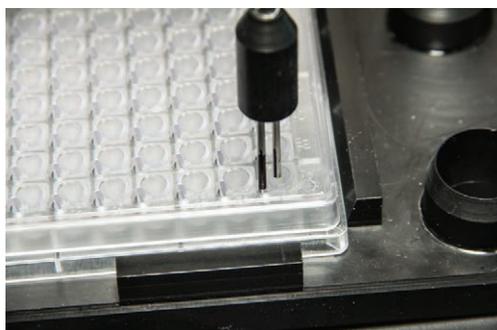


Fig. 2 — The ability of the REMS AutoSampler to precisely locate the electrode results in highly reproducible TEER measurements.

TEER measurements are stored in the computer as the electrode moves from one well to the next. The Windows-based software provides user-friendly features to acquire, display and store the tissue resistance measurements.

The **REMS** electrode is very compact and robust in design. Each of two rod-shaped probes consists of a pair of electrodes: one electrode for injecting current and the other for measuring the voltage. To take a measurement, the robot inserts one probe into the center of the filter well (apical well) and the other into the opening access slot (basal well) of the 24- or 96-well plate. **REMS** uses a switched, alternating polarity DC current to measure resistance. This provides several advantages over static DC current, including:

- Absence of offset voltages on measurements
- A zero net current being passed through the membrane means the membrane is not adversely affected by a current charge
- No electrochemical deposition of electrode metal

The **REMS AutoSampler** features two rinse station locations. The rinse station can be used to park the electrode while it is not in use. The system allows you to automate rinsing after each measurement, or you may pause the system at any time and rinse manually.

When it is parked, the voltage electrode pair are shorted together internally to reduce the possibility of an asymmetrical potential difference across the two voltage electrodes. Immersing the electrode in a saline solution with the two voltage electrodes shorted for some time provides voltage stability and a low inter-electrode potential difference. A rinse station can also provide an additional function by allowing a calibration cell with a synthetic membrane to be located in the station. It provides a quick and effective way to determine if the **REMS** electrode is functional. See “Four Corners Test” on page 23.



Fig. 3 — The REMS offers two rinsing stations.

Parts List

After unpacking, verify that there is no visible damage to the unit. Verify that all items are included:

- (1) **REMS** Robotic Sampler (vertical arm, **REMS** electrode holder, tray, tray spacers, magnetic cable support rod, power cord and RS-232 serial cable)
- (1) Computer with monitor, mouse and keyboard. The PC is pre-configured with a National Instruments PCI-6220 Data Acquisition card installed. All the National Instruments and **REMS** software is pre-installed and configured.*
- (1) **REMS** Interface Unit (RIU) with two connector cables:
 - RIU-electrode
 - RIU-PC (A/D board) in computer
- (1) Well plate holder with two rinsing stations
- (1) **REMS** Electrode(s)
- (8) **14176** Rinse Well Cups (6 may be used for electrode tests)
- (2) Test resistors (1 K Ω m and 10 K Ω m) for USER calibration or verification
- (1) **REMS** software disk
- (1) Instruction Manual

*If you are upgrading from an older **REMS** system, you may need to install new hardware and software.

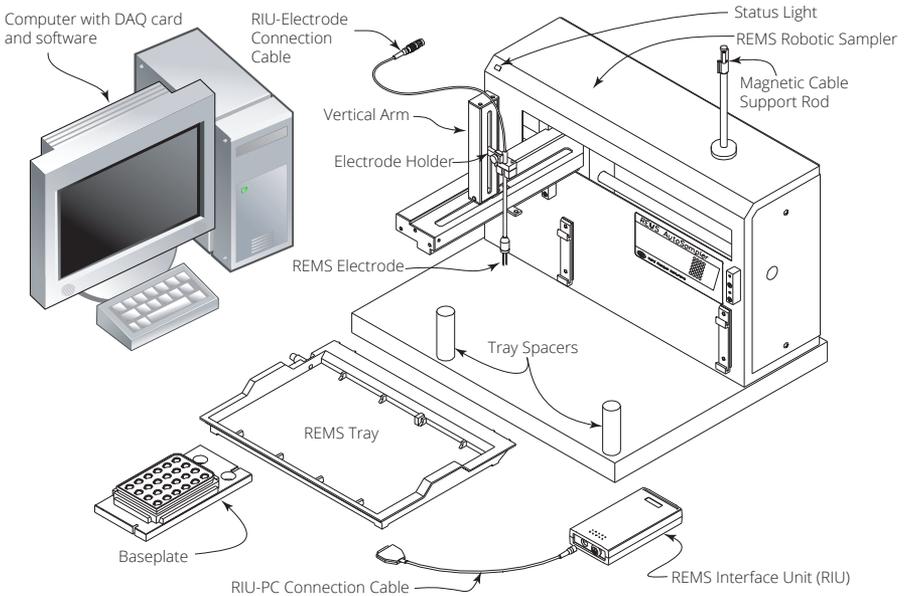


Fig. 4 — Components of the REMS AutoSampler

Unpacking

Upon receipt of this instrument, make a thorough inspection of the contents and check for possible damage. Missing cartons or obvious damage to cartons should be noted on the delivery receipt before signing. Concealed damage should be reported at once to the carrier and an inspection requested. Please

read the section entitled “Claims and Returns” on page 43 of this manual. Please contact WPI Customer Service if any parts are missing at 941-371-1003 or customerservice@wpiinc.com.

Returns: Do not return any goods to WPI without obtaining prior approval (RMA # required) and instructions from WPI's Returns Department. Goods returned (unauthorized) by collect freight may be refused. If a return shipment is necessary, use the original container, if possible. If the original container is not available, use a suitable substitute that is rigid and of adequate size. Wrap the instrument in paper or plastic surrounded with at least 100mm (four inches) of shock absorbing material. For further details, please read the section entitled “Claims and Returns” on page 43 of this manual.

NOTE: Retain the original robot packing materials. This becomes valuable if the robot ever needs to be shipped again.

INSTRUMENT DESCRIPTION

Theory of Operation

The **REMS** automates measurements of electrical resistance of transepithelial, transendothelial or CaCo-2 cell membranes being grown to confluence on microporous filters of 24- and 96-well microplates. The microplate is placed in a well plate holder filled with an electrolyte solution. The well plate holder sits on a tray on the **REMS** robotic unit. The arm of the robotic sampler moves the electrode to software selectable cells of the microplate. Resistance measurements are taken and can be stored in a text file on the computer in a standard format that can be imported into a spreadsheet program or a text file editor.

Setup

The components required for assembling the **REMS** (*Fig. 3*) include:

- Robotic sampler
- Vertical arm
- Accessory package with fuses, power cord, holder guide for electrode
- Tray and tray support posts
- RS232 serial cable

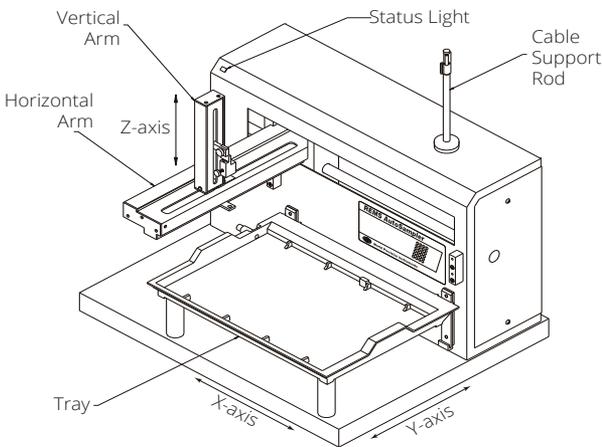


Fig. 5 — Assembled REMS

Arm Locking Screw Removal

During shipment, a screw locks the horizontal arm into place (Fig. 6).

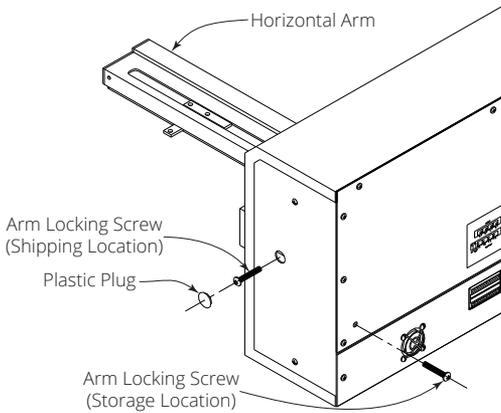


Fig. 6 — The Arm Locking Screw is located on the side panel under a black, plastic plug.

1. Locate and remove the black, plastic plug on the right side panel of the **REMS**.
2. While holding the horizontal arm in place, use a Phillips screwdriver to remove the arm locking screw, located inside the **REMS**.
3. Insert the arm locking screw into its storage location on the rear panel.
4. Replace the plastic plug on the side panel.
5. Ensure that the horizontal arm can move by pushing it to the left as far as it will go.

NOTE: Before re-packing the **REMS** for shipment or storage, always secure the horizontal arm using the locking screw. See "Packaging for Shipment" on page 28.

Vertical Arm Installation



CAUTION: Before installing the vertical arm, the power should be turned off and the power cord disconnected from the power socket. Also, check that the arm locking screw has been removed so that the horizontal arm can move.

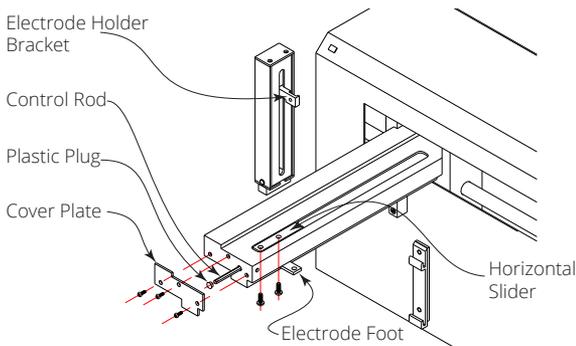


Fig. 7 — Installation of the vertical arm of the robotic sampler

1. To install the vertical arm, remove the cover plate from the front of the horizontal arm by removing its three screws (Fig. 7).
2. Locate the hexagonal-shaped control rod and horizontal slider by looking down into the horizontal arm.
3. Using your finger, press on the control rod where it passes through the horizontal slider. At the same time, pull the electrode foot or guide towards the front of the horizontal arm. This causes the white plastic plug and control rod to move forward slightly. When the white plastic plug is no longer flush with the front of the horizontal arm, remove it and the control rod from the horizontal arm.
4. Pull the electrode foot toward the front of the horizontal arm as far as it will go.
5. Position the vertical arm onto the horizontal slider. When viewed from the front, the vertical arm's electrode holder bracket is on the right.
6. With the supplied screws, secure the vertical arm to the horizontal slider. Insert the screws into the bottom of the mounting holes in the horizontal slider. The vertical arm may be required to be moved back and forth slightly to align its mounting holes with those in the horizontal slider.
7. Re-insert the control rod as far as it will go. While inserting the control rod, you may need to rotate it back and forth slightly to get it to pass through the gearing and motor drive socket in the horizontal slider.
8. Applying pressure at the vertical arm's base, push the vertical arm to the back of the horizontal arm.
9. While slightly moving the electrode holder bracket up and down, push the control rod until it clicks into position.
10. Re-insert the plastic plug.
11. Re-attach the cover plate to the front of the horizontal arm.

Tray Installation

The tray positions the well plate holder. Any liquid spills, such as those caused by the rinsing station and the overflowing of the well plate may be contained within the tray.

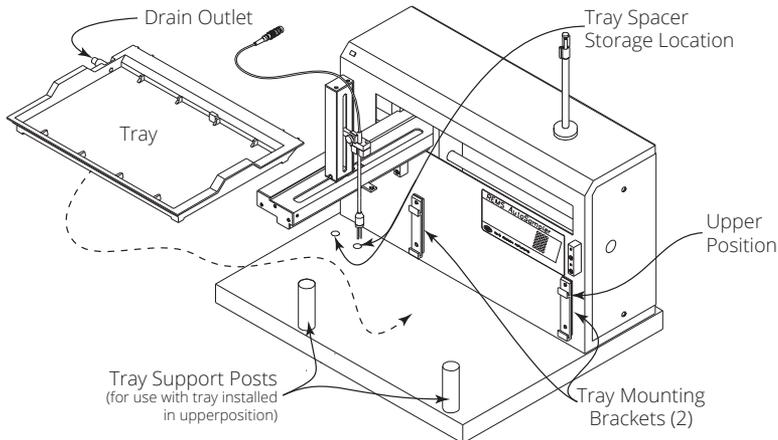


Fig. 8 — Tray installation on the robotic sampler

1. Screw the tray support posts into place (Fig. 8).

- Place the tray on top of the support posts, making sure it fits securely in the tray mounting brackets and that the drain outlet is located at the left rear of the tray.

NOTE: To remove the tray, first lift it straight up and then bring it towards you.

Installing the Well Plate Holder

The well plate holder secures the well plate in position and has two rinsing stations. It has been designed to allow for easy insertion of the well plate. Orient the well plate holder so that the end with the rinsing station is nearest to the front panel of the REMS. The slots on the well plate holder fit into the locator tabs on the tray as shown in Fig. 9.

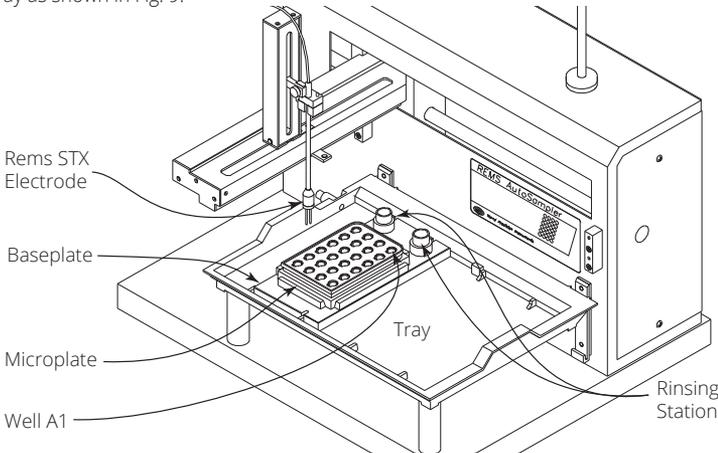


Fig. 9 — REMS AutoSampler with baseplate and REMS electrode installed

NOTE: The well plate holder has four leveling adjustment screws located at each of the four corners. These screws are adjusted at the factory so that the horizontal surface of the well plate holder is parallel to the horizontal axes of the robot. In the event that these adjustments need to be corrected, see "APPENDIX B: Upgrading from REMS 6.01 to 6.02" on page 33.

Installing the REMS Electrode Holder and Electrode

- To install the electrode holder, slide it fully into place on the electrode holder bracket and secure it with the supplied thumb screw. Fig. 10 shows the location where the electrode holder is installed on the vertical arm of the **REMS**.
- To install the **REMS** electrode, slide the electrode onto the two pegs on the electrode holder.
- Tighten the two electrode anchoring screws to hold the electrode in place. The electrode should be snug enough that it has no play, but loose enough to pop off the pegs if it hits the tray.



CAUTION: Do not over-tighten the electrode anchoring screws. These screws should be loose enough to allow a misaligned electrode to pop up off the pegs if it hits the plate instead of a well. A misaligned electrode may be damaged if it does not have some freedom of movement along the z-axis.

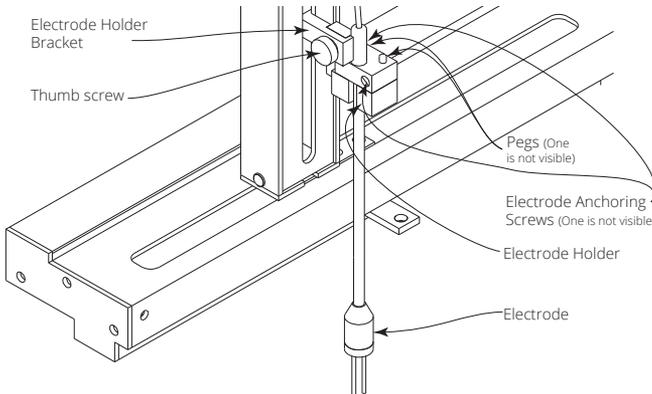


Fig. 10 — Installing the REMS electrode

Making Electrical Connections

Fig. 11 shows the rear panel of the REMS, which includes:

- RS-232 port
- Power receptacle
- Power switch
- Fuse drawer (with fuse installed)
- Switch SW1 is the unit ID selector and is preset for computer operation.
- Switch SW2 is preset for 19200 baud.



CAUTION: Switches SW1 and SW2 should be maintained at their preset positions. The other rear panel connectors are not to be used for any other purpose and may damage the REMS if used.

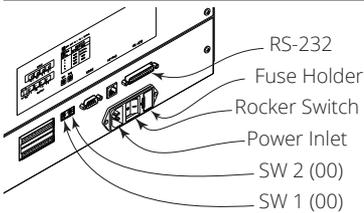


Fig. 11 — REMS rear panel

1. Be sure your computer is turned off before making any connections.
2. The RS-232 port transfers information between the **REMS** and a computer. Before operating the **REMS**, you must connect it to a computer via the RS-232 connector in one of two ways:
 - If your computer has an RS-232 port, attach the male 25-pin D-connector end of the RS-232 cable to the RS-232 port on the rear panel of the **REMS**. Tighten the retaining screws. Then, attach the female 9-pin D-connector end of the cable to the RS-232 port on your computer and tighten the retaining screws.
 - If your computer does not have an RS-232 port, you will need an FTDI serial RS-232 to USB adapter.



Fig. 12 — (Left) The FTDI Chipi-X10 is a USB to RS-232 converter cable.

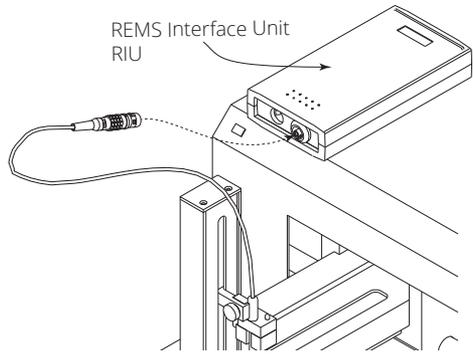


Fig. 13 — (Right) Connect the electrode to the REMS Interface Unit.

3. Connect the cable from the **REMS** electrode to rear panel of the **REMS** Interface Unit (RIU). See Fig. 13.
4. Connect the data cable from **REMS** Interface Unit to PCI-6220 data acquisition card installed in the computer.

NOTE: The system is shipped with the National Instruments software and the PCI-6220 card installed. If you need to reinstall the software or the hardware, please refer to “Appendix A: Installing the Data Acquisition Card and Software” on page 31.

5. Insert the power cord into the power inlet socket. Plug the other end into an A/C wall outlet.

OPERATING INSTRUCTIONS

Start Up

1. Ensure that the **REMS** is connected to the power source.
2. Check that cable interconnections are properly made.
3. Turn on the **REMS** robot using the power switch on the rear panel. The yellow status light on the upper left corner of the **REMS** front panel illuminates.

NOTE: A blinking status light on the far left corner of the **REMS** robot indicates a problem. See “Troubleshooting” on page 30.

Operating Precautions

Before running the **REMS** program:

- Ensure that the area around the **REMS** is free of articles that would encumber movement of the robotic arm.
- Ensure that the microplate is properly located within the baseplate and is not covered. Well A1 should be located in the far right corner. If it is loose, adjust the baseplate to secure the microplate. The rinsing stations are located on the top corners of the baseplate chamber as seen in Fig. 9.
- Arrange the cables from **REMS** electrode to avoid interference with the movement of the robotic arm.

A magnetic cable support is provided for this purpose.

- Always test the movement of the robotic arm before installing an electrode.
- A blinking status light may indicate communication with the robot has been interrupted. Reset the robot by cycling the power. If this doesn't correct the problem, see "Communication" on page 15.
- To avoid injury, take care that you are well clear of the arm.

Starting the REMS Program

The **REMS** software operates under Windows 7, 8 or 10 (32 or 64 bit). To Run the **REMS AutoSampler** program, double click on the **REMS** desktop icon.

NOTE: If the **REMS** robot is not connected, a message (Fig. 15) appears asking if you would like to run in simulation mode. Simulation mode is designed to evaluate electrodes during electrode performance testing. See "Multi-Concentration Electrode Performance Testing" on page 24. To troubleshoot the communications error, see "Troubleshooting" on page 30.

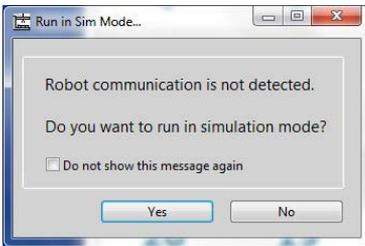


Fig. 15 — If the robot is not properly connected, this message appears.

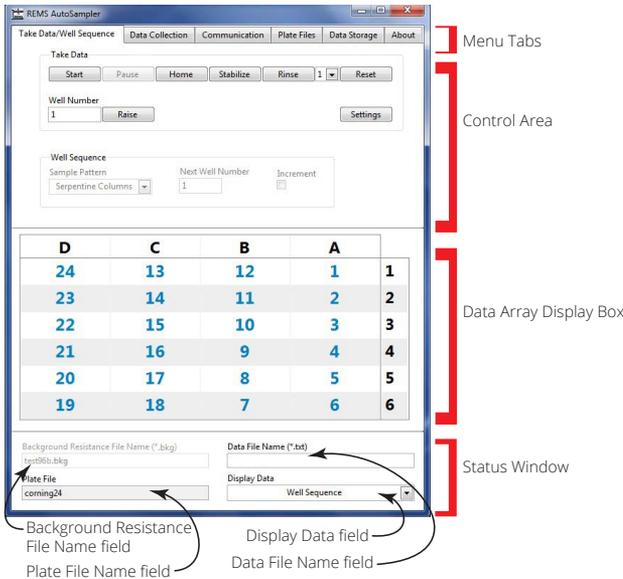


Fig. 16 — REMS System software interface

The menus are contained in the Menu Tabs:

- Take Data/Well Sequence
- Data Collection
- Communication
- Plate Files
- Data Storage
- About

REMS data is shown in the *Data Array Display Box* (Fig. 16). It displays the data selected by the *Display Data* drop down list box in the bottom right corner of the window. In the example, the well sequence is displayed. Changing the *Display Data* selection does not affect how data is taken or where it is stored. Options include: Well sequence, Well background, Total resistance and Tissue resistance.

Three fields at the bottom of the window displays the names of the files that are being read from or written to by the **REMS** program:

- **Background Resistance File Name:** The well background resistance is the blank resistance of the well plate and is taken before cells are cultured. This data is later used to calculate the tissue resistance by subtracting the well background resistance from the total resistance after cells are cultured. When this field is active, measurements are stored as a background resistance file (.bkg). Type a new name into the field to create a new well background resistance data file. These files are stored at C:\Users\Public\Documents\REMS\Plate_Background\. You have the opportunity to redirect these files to your own documents folder (C:\Users\[user name]\Documents\REMS\Plate_Background\).
- **Data File Name:** The total resistance measurements, which are taken after cells are cultured, is stored in the data file. By selecting this field, *Total Resistance* is automatically selected as the *Resistance Type*. If you prefer, you may select *Total Resistance* as the *Resistance Type* on the *Data Collection Setup* tab to accomplish this same result. This field is grayed out when *Total Resistance* is selected as the *Resistance Type*. To change the name of the data file, simply enter a new name into this text field when the field is active. The file is stored in .txt format, and is typically locate in C:\Users\Public\Documents\REMS\Plate_Data\. You have the opportunity to redirect these files to your own documents folder (C:\Users\[user name]\Documents\REMS\Plate_Data\).
- **Plate File Name:** The robot uses this file to position the electrode. See “Plate Files” on page 16. This field is read-only.

NOTE: By default the **REMS** files are stored in the C:\Users\Public\Documents\REMS\Plate_Configurations folder. For multiple users who log in to the **REMS** PC under different profiles, the files in this directory structure are stored in a common location where all users may access them. WPI recommends leaving this pointer at the public location in case there is an X-Y-Z change to a plate file. Then all users access the most recent plate file.

NOTE: Each user's documents directory will have the **REMS** folders copied over on the initial running of the **REMS** software, including the plate configuration files.

Understanding the Interface

The tabs across the top of the **REMS** program window are used as follows:

- **Take Data/Well Sequence**– Start/stop/pause the robotic operations. Set the sample pattern used. See “Taking Data/Well Sequence” on page 12.
- **Data Collection** lets you define the type of data collected (either background resistance or live cell measurements, whether the electrode is rinsed between measurements, and the electrode location when measurements are completed. See “Data Collection” on page 14.

- **Communication** lets you check the status and settings of communications with the robot and the data acquisition card. See “Communication” on page 15.
- **Plate Files**—Add, edit or delete plate setup files which define the coordinates for the robot’s movements. See “Plate Files” on page 16.
- **Data Storage**—Establish how data is saved or open a saved file. See “Data Storage” on page 18.
- **About** allows you to see version information. See “About” on page 19.

Taking Data/Well Sequence

This dialog box is divided into two sections. The first section (Take Data) contains the controls for starting and stopping the robotic sampling process. The second section (Well Sequence) lets you define the sampling sequence.

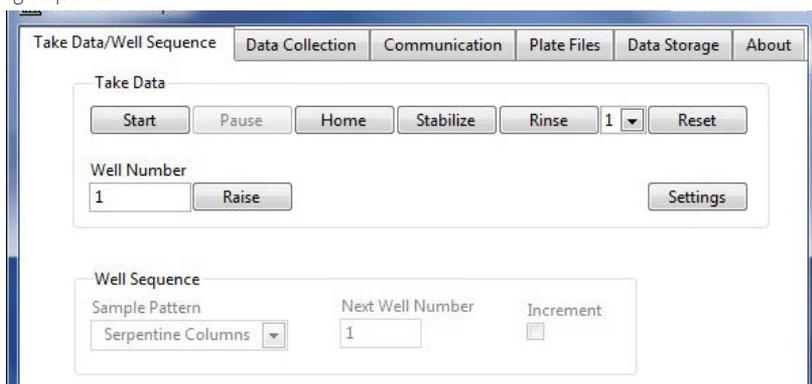


Fig. 17 — Take Data Tab

Take Data

Start: This button starts/stops the **REMS** plate measurement sequence. When **Start** is clicked, its name changes to **Stop**. Clicking on **Stop** halts the robotic arm at its current position. The **REMS** restarts sampling from the beginning of the sequence (default: well A1) when **Start** is pressed again.

Pause: This button stops the data collection process initiated with **Start**. This is useful for double-checking or changing parameters before continuing with TEER measurements. Press **Start** to resume sampling with the next well in your sequence.

Home: Returns the robotic arm to the home position. The home position is located at the maximum limit of the robotic arm’s travel, in the upper, left-hand, rear corner.

Stabilize: When this button is pressed the electrode moves to the user-defined location for the stabilizing well as defined by the settings in the *Plate File Settings Window* (Fig. 26 on page 17). The *Plate File Settings Window* is accessed from the *Plate Files* tab. A resistance measurement is displayed each time this button is pressed.

Rinse: REMS AutoSampler is equipped with two rinsing stations. Click this button to move the electrode to the rinse station indicated by the drop down list next to the rinse button. In the setup shown in Fig. 17, the electrode parks in Rinse Station 1 when you press **Rinse**. No resistance measure is displayed when this button is clicked.

Raise: This button becomes active after a stop or pause command. Press this button to raise the electrode to its highest position while maintaining its X-Y coordinates.

Reset: This button clears the data array of all the microplate resistance data, including the well background and total resistance. Unsaved data is deleted. Pressing this button does not affect the well sampling sequence.

Well Number: This read-only value indicates the well position of the electrode.

Settings: Press this button to see a snapshot of the current data collection and storage settings (Fig. 18). Pressing **See Procedure** on this window brings up a short summary of what is happening on the active read (Fig. 19).

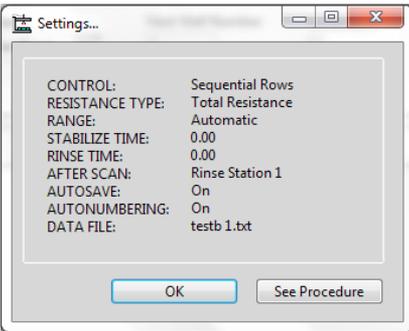


Fig. 18 — When you press the Settings button, a window displays showing a summary of your current setup.

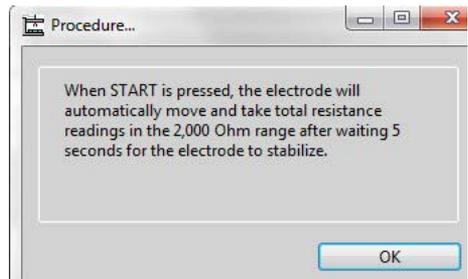


Fig. 19 — The Procedures window describes the programmed action.

Well Sequence

Use the Sample Pattern drop down list (Fig. 21) to define the order in which the wells are sampled. There are five sample patterns, four of which are presets and one which is user-defined.



Fig. 21 — Choose a sample pattern.

- **Sequential Rows:** A1, A2, A3, A4, A5, A6, B1, B2...
- **Sequential Columns:** A1, B1, C1, D1, A2, B2...
- **Serpentine Rows:** A1, A2, A3, A4, A5, A6, B6, B5...

- **Serpentine Columns:** A1, B1, C1, D1, D2, C2...
- **User Defined** lets you define a custom sequence. In order to create your own sequence, first select *Clear Sequence* from the list to set everything to zero and clear existing patterns. Then, click on each cell in the table to define the sequence. The cells will be sample in the order of your selections. The *Next Well Number* field displays the next number in your sequence. If the *Increment* option check box is selected, the *Next Well Number* automatically increments each time a cell is added.
- **Clear Sequence** sets all cells in the sequence array to zero. Do this prior to creating a new user-defined sequence.

Data Collection

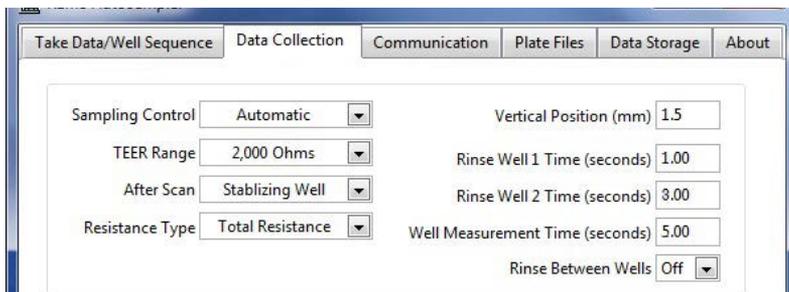


Fig. 22 — Data Collection Tab

Sampling Control: Choose either *Automatic* or *Manual*. In *Automatic* sample mode, the **REMS** samples in the sequence specified in the *Sample Pattern* drop down box. In *Manual* sample mode, the **REMS** samples the cell that you click on.

TEER Range: This drop down list sets the resistance range the **REMS Autosampler** measures. Range options include *Automatic*, *20K Ω* and *2K Ω* . If *Automatic* is selected, then the range automatically switches to the appropriate range.

After Scan: Select where the **REMS** electrode will located after the microplate sampling sequence is complete. Options include:

- Remain in the last sampled well (*Remain in Well*)
- Go to the robot home position (*Home Position*)
- Go to the stabilizing well (*Stabilizing Well*)
- Go to one of the rinse stations (*Rinse Station 1* or *Rinse Station 2*)

Resistance Type: This drop-down list selects the data file type that will be created when data is saved (total resistance or well background). Before any tissue is grown on the HTS plate, a blank well reading should be taken and saved as a background file type. Later, after cells are cultured, the total resistance measurements are taken. The background resistance is automatically subtracted from the total resistance measurements to yield the actual resistance of the tissue.

NOTE: When *Total Resistance* is selected, the *Data File Name* text field in the status window becomes active, and the *Background Resistance File Name* text field is grayed out (Fig. 23). This means that data will be written to the indicated file type when measurements are saved, and data in this file is displayed in the *Data Display Array* box. Conversely, when *Well Background* is selected, the *Data File Name* field is grayed out and the *Background Resistance File Name* text field is active. This means that the indicated background data file will be written to when **REMS** data is saved. To change the name of either file, simply enter a new name into the appropriate text field when the field is active. Data is written to the new file when a save action is executed.

(On the Data Storage tab, press [Save Data](#))

Background Resistance File Name (*.bkg)	Data File Name (*.txt)
MP_BG_08 .bkg	150mM_p6.txt
Plate File	Display Data
96MILIPORE_TM	Well Background

Fig. 23 — When Total Resistance is selected as the Resistance Type (on the Data Collection tab - page 14), the Background Resistance File Name field is grayed out, and the Data File Name field is active.

Vertical Position: This field determines the electrode's lowest position in the well when a sequence is executed. This number represents the distance in millimeters above the Z Low position defined on the Plate File Settings tab in the Position of Well A1 section. The A1 Z Low position is typically set to correspond to the electrode tip almost touching the membrane. Values may be entered to the nearest 0.1mm. Usually, this is set to 1-2mm.

Rinse Well (#) Time: Indicate the time in seconds that the electrode remains in the rinse station between measurements.

Well Measurement Time: This field indicates how long the electrode remains in a well taking a measurement.

Rinse Between Wells: Choose whether to rinse the electrode between measurement and which of the two available well locations is used. Options include:

- Off—Electrode is not rinsed between measurement
- 1—Electrode rinses in Rinse Well 1 only
- 2—Electrode rinses in Rinse Well 2 only
- 1&2—Electrode rinses in both rinse wells

Communication

Take Data/Well Sequence	Data Collection	Communication	Plate Files	Data Storage	About										
<table border="1"> <tr> <td>NI DAQ Device</td> <td>NI DAQ Device Status</td> </tr> <tr> <td>Dev1</td> <td></td> </tr> <tr> <td>Robot Serial Port</td> <td>Robot Serial Port Status</td> </tr> <tr> <td>COM1</td> <td>Robot failed to connect</td> </tr> <tr> <td>Reset Robot</td> <td></td> </tr> </table>						NI DAQ Device	NI DAQ Device Status	Dev1		Robot Serial Port	Robot Serial Port Status	COM1	Robot failed to connect	Reset Robot	
NI DAQ Device	NI DAQ Device Status														
Dev1															
Robot Serial Port	Robot Serial Port Status														
COM1	Robot failed to connect														
Reset Robot															

Fig. 24 — Communication Tab

NI DAQ Device Number: The REMS system uses a data acquisition card manufactured by National Instruments. The device number is set at the factory and does not need to be changed. For more information, see "Appendix A: Installing the Data Acquisition Card and Software" on page 31.

NI DAQ Device Status: The text area shows the status of the NI DAQ board. If errors exist, they will display here. See "Troubleshooting" on page 30. This is normally blank.

Robot Serial Port: This drop down box indicates the COM port on the computer that the robot uses for communications. This is set at the factory.

Robot Serial Port Status: One of three messages displays in this text area, including:

- Robot connected—No action is required.
- Robot failed to connect—Verify that the robot is turned on and the cables are securely connected. Then, press .
- Serial device not preset or accessible—Press .

If an error message persists, refer to “Troubleshooting” on page 30 or call technical support at 941-371-1003.

: If communications settings get changed, it may be necessary to re-initialize the robot by pressing this button.

Plate Files

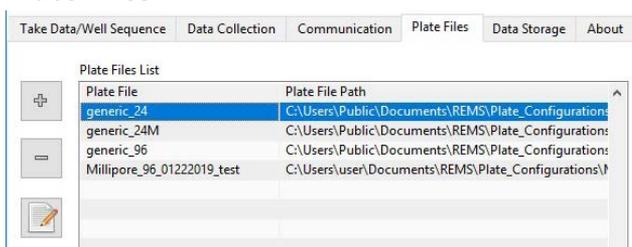


Fig. 25 — Plate Files Tab

Plate Files List: This list shows the names of the plate files that are immediately available for selection (Fig. 25). The highlighted file is the one that is currently loaded in the system. The selected file appears in the *Plate File* field in the bottom left corner of the **REMS** status window. This list only displays the plate files that have been added to the *Plate File List* from the plate configuration file folder.



The add button opens the plate configuration folder where you may choose a file to include in the *Plate Files List*.



Highlight a file in the *Plate Files List* and click this button to remove the plate configuration file from the *Plate Files List*. The file is not deleted from the plate configuration folder.



Highlight a file in the *Plate Files List* and click the edit button to bring up the *Plate File Settings* screen where you can edit and save the plate configuration file. Don't forget to save any changes made to the file before exiting.

The plate files contain all the dimensional parameters for a well plate, as well as the location data for the rinse wells and stabilizing well. These files are added from the **REMS** folder to the *Plate File List* where they are available for loading as the “active” plate file.

Plate File Settings



Highlight a file in the *Plate Files List* and click the  button to edit the file. The *Plate File Settings* window appears (Fig. 26). You have the option of saving the file under a new name, if you want to create a new plate configuration file. This screen provides functions for creating, editing and saving plate configuration files. It also permits manual control of the robot for testing purposes. To setup parameters for a new plate, see “Setting Position Parameters for a New Plate” on page 21.

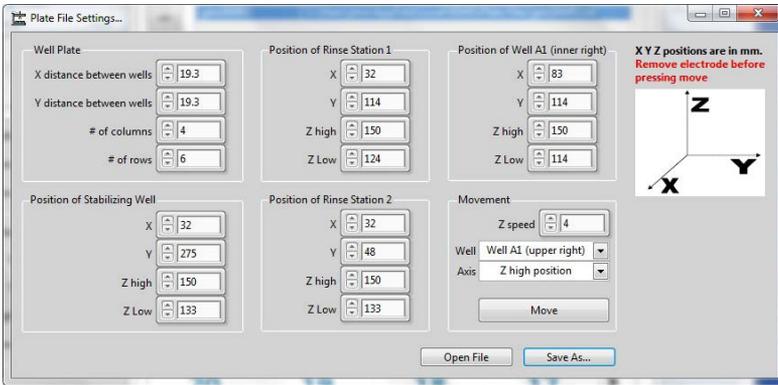


Fig. 26 — REMS Plate File Settings window lets you edit plate configuration files.

Open File

: Opens the folder where plate configuration files are stored so that you can select an existing plate file to edit.

Save As...

: Save the modification to a new plate file or overwrite the existing file.

It is advised to home the electrode and test the A1 position a few times to assure the position. (Save. Home. Open File. A1. Adjust if necessary. Save. Repeat.)

Well Plate Section

X distance between wells: This field sets the X distance between the wells in 0.1mm increments.

Y distance between wells: This field sets the Y distance between the wells in 0.1mm increments.

of columns: This field sets the number of columns in the tray. It is typically set to 4 for the 24-well plate and 8 for the 96-well plate.

of rows: This field sets the number of rows in a tray. It is typically set to 6 for the 24-well plate and 12 for the 96-well plate.

Position of Well

Four locations of the electrode are specifically defined by their coordinates. These include:

- **Stabilizing Well**—The fields in this section set the position the robot moves the electrode to when it goes to the stabilizing well.
- **Rinse Stations 1 and 2**— The fields in this section set the position the robot moves the electrode to when it goes to each of the rinse stations.
- **Well A1 (upper right corner)**—The fields in this section set the position the robot moves the electrode to when it goes to the upper right corner well. (This refers to the well closest to the robotic sampler on the right side when you are facing the robotic sampler. See [Fig. 9](#) on page 7.)

X: This field sets the X coordinate of the electrode position.

Y: This field sets the Y coordinate of the electrode position.

Z high: This field sets the upper (vertical) Z coordinate of the electrode position above the well. The Z high position should be set to a position higher than the well plate, because this is the elevation at which the electrode will travel when moving between wells.

Z low: Typically this is set to the lowest point in the well (just touching the membrane) as a reference point. This is not necessarily the low position of the electrode during a sampling sequence. The Z low electrode height (during the execution of a sequence) is set relative to the setting defined here, as defined in the *Vertical Position* field in the Data Collection tab (Fig. 22 on page 14).

Movement

Z speed: This sets the speed of the Z-axis movement. Valid settings are 1 through 5. The default setting is 5. Slower settings are useful to visualize the movement of the electrode position changes. This setting does not affect the speed of the robot movement when a sequence is executed.

Well: This control selects which positional data set is referred to when the  button is pressed. The options are *Well A1*, *Rinse Station 1*, *Rinse Station 2* or *Stabilizing Well*.

Axis: Selects the axis that the robotic sampler moves on when the  button is pressed. The options are *XY position*, *Z high* or *Z low*.

: Press this button to move the electrode to the position designated by the *Well* and *Axis* fields. The electrode always moves to the Z-high position before making any lateral movements.

 **CAUTION:** Make sure the Z-high position is programmed above the highest landmark on the well plate.

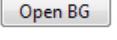
Data Storage

From this window, you can open and save background resistance files and data files. The well background resistance measurements are stored in the background resistance files (.bkg files) in the **REMS** directory. The data file contains the well identifier, total resistance measurements, well background resistance, tissue resistance and well sequence information. These files are typically located in C:\users\Public\Documents\REMS\ **REMS** data arrays (.txt files) are recorded as tab delimited text files. This format has the advantage of being compatible with both word processors and spreadsheet programs like Microsoft® Excel.

NOTE: The background data file has a .bkg extension, and the data file has a .txt file extension to distinguish between the two file types. They are stored in separate folders (directories) inside the REMS folder.



Fig. 27 — Data Storage Tab

 : Use this button to open an existing **REMS** data file (or load a well background file). If *Total Resistance* is selected as the *Resistance Type* on the *Data Collection* tab,  displays. If *Well Background* is selected, then  displays.

 : Use this button to manually save the data shown in the *Data Array Display* box in the location designated by the *Data Folder* or *Background Resistance Folder* fields. If *Total Resistance*

is selected as the *Resistance Type* on the *Data Collection* tab, **Save Data** displays. If *Well Background* is selected, then **Save BG** displays.

Auto Save: Check this box to automatically save the data when the **REMS** system has completed a sequence. If the *Auto Numbering* checkbox is not selected, the previous data file is overwritten, unless it is manually re-named before saving.

Auto Numbering: Select this checkbox to append the data file name with the integer that appears in the adjacent “#” field. The integer value increments automatically when a file is saved. That way, the new data does not overwrite existing data files.

NOTE: The *Auto Numbering* fields only appear when *Total Resistance* is selected as the *Resistance Type*. Background data files must be named manually.

Data Folder: This editable text field indicates the path and folder location where the data files for the TEER measurements are stored. The name of the active data file is set in the *Data File Name* field in the status window which appears at the bottom of the screen (Fig. 16) regardless of which tab is selected. You have the opportunity to redirect these files to your own documents folder (C:\Users\[user name]\Documents\REMS\Plate_Data\). The default location is C:\Users\Public\Documents\REMS\Plate_Background or Plate_Data.

Background Resistance Data File Path: This editable text field indicates the path and folder location where the data files for the well background resistance measurements are stored. The name of the active background resistance file is set in the *Background Resistance File Name* field in the status window at the bottom of the **REMS** window.

NOTES: Enter any notes about the plate or the experiment that you want to store in the data file.

About

Click the **About** tab to find the **REMS** version information.



Fig. 28 — The *About* tab shows the version of your *REMS* software.

Getting Started

This procedure assumes that you have a proper plate file pre-configured. An appropriate plate configuration file should have been provided by WPI. If you do not have a configured plate file, see “Setting Position Parameters for a New Plate” on page 21.

1. Setup the hardware. See “Setup” on page 4.
2. Verify that the tray, well plate holder and other hardware are correctly located and oriented on the **REMS**.
3. Fill the well baseplate chamber with perfusate (or saline) solution.
4. Insert the microplate without the cultured tissue into the baseplate chamber. The well resistance measurements taken in this condition corresponds to the background resistance.
5. Boot the computer. Start the **REMS** software by double clicking on the desktop icon.

NOTE: REMS saves the last setup you were working on when you close the program. Each time the program launches, it opens to the data files used when you last closed it.

6. Select the *Plate Setup* tab, and choose the correct plate configuration from the *Plate File* list. See “Plate Files” on page 16.



CAUTION: If the wrong plate file is selected, the probe could be damaged. It is always best to test the plate before an actual run is performed.

7. Set the other parameters as desired on the *Take Data/Well Sequence* and *Data Collection Setup* tabs. See “Taking Data/Well Sequence” on page 12.
8. Select the *Data Storage* tab, and choose the storage location for the *Background Resistance Data File*. To change the folder location, click  next to the *Background Resistance Data File* field and navigate to the folder where you wish to store the background file. Click **Current Folder** to set the current location as the default storage file location. Do NOT enter a filename. If you want the file to save when the measurement run completes, check the *Auto Save* button. Select the *Auto Numbering* checkbox to automatically increment the file numbering so that you never overwrite an existing data file.

NOTE: To open an existing background file, click **Open BG**. Highlight the background file of choice and click **OK**. The background file name is loaded as the active background file in the status window (Fig. 16 on page 10).

9. Select the *Take Data/Well Sequence* tab, and press **Stabilize**. The electrode should move to the stabilizing well. Allow the electrode to equilibrate to at least 15 minutes to stabilize. Equilibration can also be accomplished in the Well A1.
10. Select the *Data Collection Setup* tab, and set the *Resistance Type* to *Well Background*. The *Background Resistance File Name* field at the bottom of the window becomes active.

11. Select the *Take Data/Well Sequence* tab, and press **Start** to initiate resistance measurements. The **Start** turns into **Stop**. If you need to stop the robot, you can press **Stop**.

The **REMS** starts the sample sequence from the beginning when **Start** is pressed again. To temporarily stop the data collection process, press **Pause**. This is useful for double-checking or changing parameters before continuing with TEER measurements. If the sampling is paused, press **Start** to resume sampling at the next well.

12. At the end of the run, the **REMS** electrode does what you designated on the *Data Collection Setup* tab in the *After Scan* drop down list. It could remain in the well, go to the stabilizing well, or return to the home or one of the rinse well positions.
13. Select the *Data Storage* tab. If *Auto Save* was not selected, be sure to save the file by pressing the **Save BG** button. The file is stored in the location established in the *Background Resistance Folder* field. If a file by the designated name already exists at that location, a warning appears.

To overwrite, click **OK**. Otherwise, click **Cancel** and enter a new name in the *Background Resistance Data Folder* field, and then save the file.

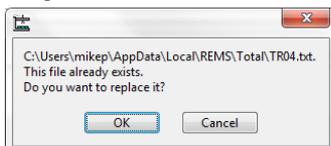
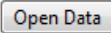
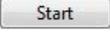
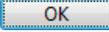


Fig. 29 — A warning appears if you try to overwrite an existing file.

14. When you are ready to measure the resistance of a cultured monolayer, select the *Data Collection Setup* tab, and set the *Resistance Type* to *Total Resistance*. The *Data File Name* field at the bottom of the window becomes active.
15. Verify that you are using the correct plate file. The *Plate File* field in the status window indicates the plate file that is loaded. This file defines the position of the wells and controls how the robot moves. To choose a different plate file, select the *Plate Setup* tab and choose the correct plate file. See "Plate Files" on page 16.
16. Select the *Data Storage* tab, and enter the storage locations for the data file. To select a file location, click  next to the *Data Folder* field and navigate to the folder where you wish to store the file. Click  to set this folder as the current storage location. If you want the file to save when the measurement run completes, check the *Auto Save* button. Select the *Auto Numbering* checkbox to automatically increment the file numbering so that you never overwrite an existing data file. This does not create any files. Rather, it establishes the storage location.

NOTE: To open an existing data file, click . Then, navigate to the folder, highlight the file and click .
17. To view the calculated tissue resistance, select *Tissue Resistance* from the *Display Data* drop down box at the bottom of the **REMS** window.
18. Select the *Take Data/Well Sequence* tab, and press the  button to initiate resistance measurements. The  turns into . If you need to stop the robot, you can press . The **REMS** resumes sampling at the next well when  is pressed again. Allow the robot to finish making measurements.
19. Select the *Data Storage* tab. If *Auto Save* was not selected prior to the run, be sure to save the file by pressing . The file is stored in the location established in the *Data Folder* field. If a file by the designated name already exists at that location, an overwrite warning appears. To overwrite, click . Otherwise, click  and enter a new name in the *Data Folder* field, and then save the file.
20. Repeat steps 14-20, as needed.

Setting Position Parameters for a New Plate

A Plate File is comprised of a data set that defines the locations of the wells on a microplate. It also defines the location of two optional rinse stations and a stabilization well which are usually positioned off the microplate footprint. Pre-configured plate files are provided by WPI for the most popular manufacturer's models in either 24 or 96-well count. You can also create your own plate files. On occasion, you may find it desirable to make minor adjustments to a factory preset.

The easiest way to create a new plate file is to edit an existing plate file. The data points are defined in the *Plate File Setting* window. Only eight data points are required to define all the positions on a microplate. These eight data points are defined in the fields within the *Well Plate* and *Position of Well A1* sections of the *Plate File Settings* window. To define new coordinates for robot movement, click on the *Plate Files* tab, highlight any

plate file to edit and press  to open the *Plate File Settings* window and save it with a new name.

1. Before making any adjustments, remove the electrode from robotic arm by lifting it straight up. (You may have to loosen the two set screws on the top of the electrode first.) See "Installing the REMS Electrode Holder and Electrode" on page 7.

2. Install the microplate in the microplate holder on the **REMS AutoSampler** stage.
3. Select the *Plate Files* tab. To edit an existing plate file, highlight the file in the *Plate File List* and press .

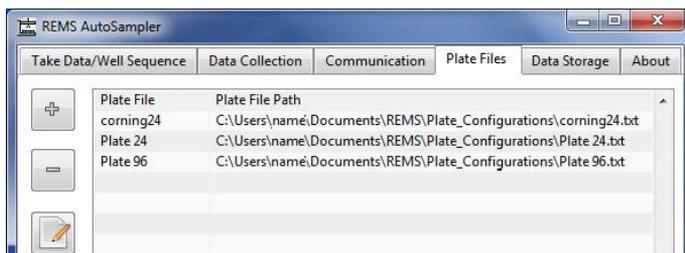


Fig. 30 — To edit an existing plate file, highlight the file and press the Edit button.

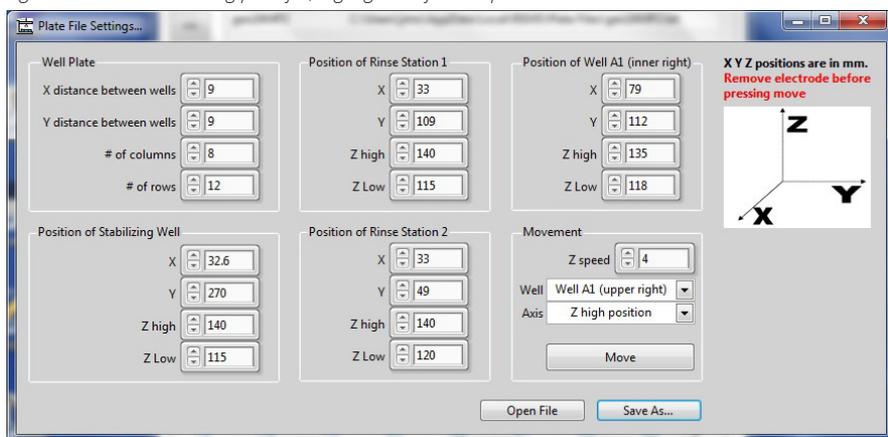
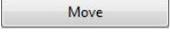
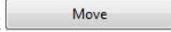
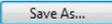


Fig. 31 — Plate File Settings window

4. In the *Movement* section (lower right hand corner of the window) select *Well A1*. Select *XY Position* from the *Axis* drop down list.
5. Verify that the electrode has been removed. It is safest to run the movement protocol without an electrode the first time to avoid crashing an expensive electrode. Press . The robot arm moves to the designated location for the *A1 well*.
6. On the *Plate File Setting* window in the *Movement* section, change the *Axis* drop down list to the *Z-low* position and click .
7. Install the dummy electrode to see if the probes are centered over the apical well opening and basal access port.
8. At this time, raise the *Z low* position so that the electrode barely protrudes into the well. This makes visualization of the *X-Y* coordinates during the initial test run much easier.
9. Check the electrode position with reference to the well. The electrode should be positioned in the center of the opening. If the electrode is not centered, change the *Axis* to *X-Y Position*, and adjust the *X* and *Y* coordinates. Click  again to move the electrode in small increments to redefine the electrode position above the *A1 Well*.

NOTE: The XYZ diagram (located in the upper right corner of the *Plate File Settings* window) shows the orientation of the X, Y and Z axes when the operator is facing the front of the robotic sampler. The arrows point in the positive direction.

- Before continuing, save the plate file by clicking  and entering a new name for your plate file. Click . It is advised to check the A1 position (XY) by homing the electrode, opening the plate file and testing the A1 position a few times to assure proper positioning. (Save. Home. Open File. A1. Adjust if necessary. Save. Repeat.)
- Measure the distance from center to center of adjacent wells in both the X and Y directions. Verify that they match the values in the *Well Plate* section of the *Plate Files Settings* window.
- On the *Take Data/Well Sequence* tab, press  to run the sequence. Verify that the electrode positions itself in the center of each well as it moves through the sequence.
- Open the *Plate Files* tab and click  to open the *Plate Files Settings* window.
- Adjust the Z-Low position of the electrode until the electrode almost touches the membrane. It should be positioned about 1-2 mm above the membrane.
- Use a similar alignment procedure for setting up the locations of the rinse stations and stabilizing well.

NOTE: The stabilizing well is unique in that it allows you to see a resistance measurement when the electrode samples that well. You may program the stabilizing well with the same coordinates as one of your rinsing wells, if you prefer.

Four Corners Test

Running an entire sequence is a comprehensive test, which may not always be required. When switching between plate type, you may choose to run a four corner test instead. This test uses a custom pattern that samples the wells at the four corners of the plate.

- After setting the X and Y coordinates on the *Plate File Setting* window, set the Z-Low value so that the probe is positioned 1–2mm above the plate.
- Save the file and close the *Plate File Setting* window.
- Select the *Data Collection* tab. Set the *Sampling Control* value to *Manual*.
- Select the *Take Data/Well Sequence* tab. Set the *Sample Pattern* to *User Defined*.
- Press .
- In the Data Array Display area, left click on each of the four outermost corners of the plate (Fig. 32) to verify that the electrode is properly centered over each of the four corner wells.

D	C	B	A	
24	13	12	1	1
23	14	11	2	2
22	15	10	3	3
21	16	9	4	4
20	17	8	5	5
19	18	7	6	6

Fig. 32 — Select the four corners in sequence to test the electrode positioning.

- When the test run has completed, press .

8. If necessary, select the *Plate Setup* tab, and edit the plate configuration file.
9. If you need to edit the plate configuration file again, repeat the test again.

Multi-Concentration Electrode Performance Testing

This procedure uses different concentrations of KCl which have been empirically tested to yield repeatable resistance values. The response of the electrode should be linear for the middle concentrations, but on the upper and lower ends of the concentration spectrum the values may deviate from linearity.

1. Prepare a stock solution of 320mM KCl (WPI# **REMS-CAL**). From this, perform dilutions to create lower concentrations of 160mM, 80mM, 40mM, 20mM and 10mM. A minimum volume of 5mL of each solution is required to perform the test.
2. Fill (6) 5mL vials (WPI #**14176**). Dispense 5mL of each concentration into each of the six vials. It might be useful to label each of the vials with the concentration to avoid confusing them when performing the test (Fig. 33).

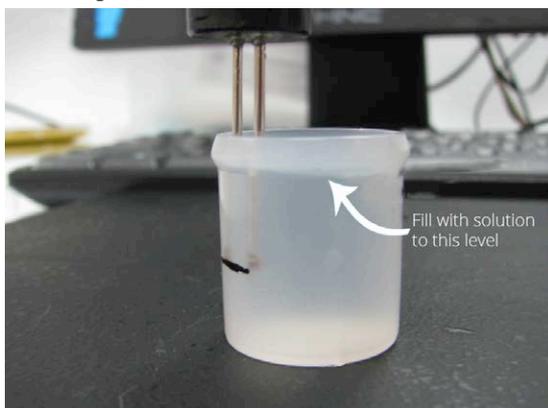


Fig. 33 — The vial should be filled to a volume of 5mL.

3. Prepare a small container of distilled water for rinsing the electrode when switching between concentrations.
4. Plug the electrode connector into the REMS electronic module.
5. Mount the electrode into a suitable holding device that will allow repeatable vertical positioning of the tip in the test vials. An economy micro-manipulator mounted to a magnetic stand with the X-axis oriented vertically works great for this purpose. A picture of the setup is shown on the next page (Fig. 34).

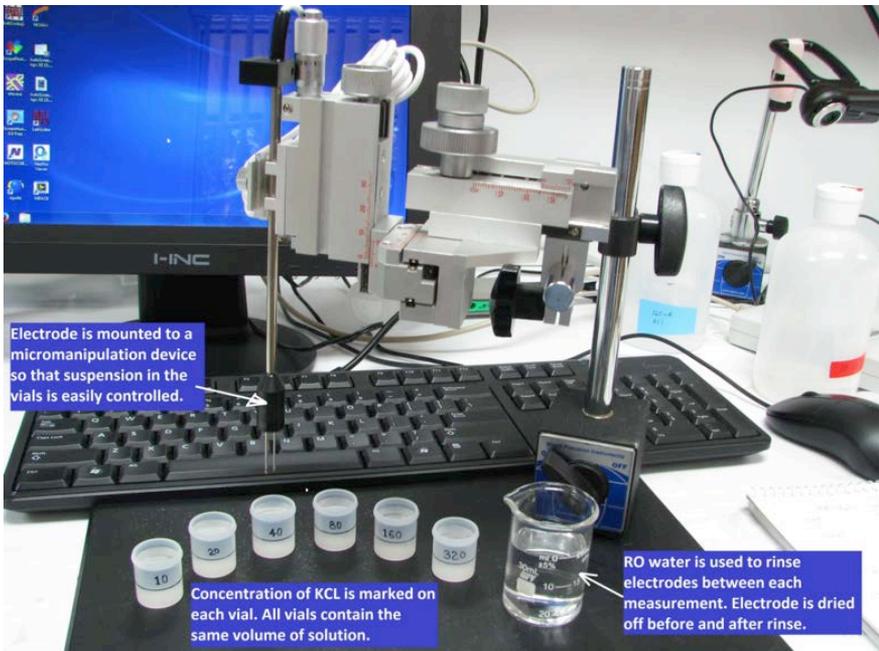


Fig. 34 — Setup for performance verification using concentrations of KCl.

6. The electrode should be tested by beginning measurements with the solution of lowest concentration first and incrementally working through the set in order of increasing concentration. For each test run, the electrode is positioned in the center of the vial, immersed to a depth of 1 cm below the edge of the lower lip of the vial.
 7. Immerse the electrode into the first solution (10mM KCl) and let it soak for 10-15 minutes.
 8. Power on the computer and launch the **REMS** software. There is no need to power up the robot, because it is not required to run the test. Run the software in "simulation mode" to eliminates the robotic movement sequences since the electrode is not mounted on the robot arm. The procedure to run in simulation mode is explained in detail in step 11.
 9. Select a plate file to run the test. The plate file is selected from the *Plate Files* tab ("Plate Files" on page 16) by highlighting the file with a mouse click. Typically a 24-well plate file is selected. A 96-well plate file takes more measurements and requires more time.
 10. Save all test run measurements as Background Resistance Files, because this is the most basic data file composed of a single data array. To record a background resistance measurement, click on the *Background Resistance File Name* field to activate it as the file type to be saved (as described on page 13 in the manual). Type a file name into the field. The file name should contain relevant basic information, which might include the model of the electrode and the concentration used for the test run. At the end of the run, this file will either be automatically saved or can be manually saved. To save files automatically, click on the *AutoSave* checkbox located under the "Data Storage" tab. (See "Data Storage" on page 18). Alternatively, you can choose not to save the file, and just record the average of the measurements taken for a given concentration value.
- If you choose the *Autosave* routine, each successive run is saved as the name that appears in the *Background Resistance File Name* field at the end of the run automatically. To avoid overwriting a previously recorded file with the same name, type a new name into the *Background Resistance File Name* field PRIOR to executing the run.

- In the lower right hand corner of the *Status Window* (Fig. 16), be sure to select *Background Resistance* as the display selection so that you can observe the data as it is being collected.
- Go to the *Take Data/Well Sequence* tab, and click on in the upper left hand corner. A message appears saying that "Communication with the robot is not detected. Do you want to run in simulation mode?" Click Yes.
- Click again. The electronic module executes a run, and you should observe resistance values being written onto the *Data Array Display Box* (Fig. 16).
- When the run is complete, you can choose to save the file or not. You may not wish to save the file if you are just checking performance and don't care to document it in a saved file. In this case, un-check the *Autosave* checkbox located under the *Data Storage* tab. If you want to save the file and the *Autosave* checkbox was not selected, manually save the file by clicking on the button under the *Data Storage* tab. See "Getting Started" on page 19 (step 13).
- Raise the electrode out of the test solution and gently dry it off with a Kimwipe or lint free absorbent paper. Then, immerse the tips of electrode into the rinse cup of distilled/RO water for 10-15 seconds. Remove the electrode from the distilled/RO water and wipe it dry once more. Use caution when drying the electrodes, because they are somewhat delicate.
- Position the next concentration vial under the electrode and lower the tips down to the 1 cm mark on the vial. Then, center it. Click the button to re-initialize the display, and then click the button. Measurements can be taken immediately since the electrode has been pre-conditioned from the 10-15 minute soak. You may notice that the resistance measurements decrease incrementally as it goes through the plate cycle. You may choose to run through the cycle several times until measurements stabilize.
- Use the table below to compare your resistance measurements obtained to the average values recorded at the factory using new electrodes at room temperature.

Test values should fall within $\pm 10\%$ of the running average. In the event any of the values falls outside the 10% range, please contact Technical Support at (941) 371-1003.

REMS Electrode Running Averages

KCl [mM]	REMS-96C [Ω]	REMS-96 [Ω]	REMS-24 [Ω]
10	385	414	515
20	203	224	276
40	103	113	141
80	54	60	73
160	29	32	39
320	16	16	21

Testing the REMS Electronics

The **REMS** system includes two test resistors, 1.0 k Ω and 10 k Ω . These resistors are effectively "dummy" electrodes that are used to verify that the **REMS** electronics are functioning correctly.

- Plug the 1.0k Ω resistor into the electrode connection port on the rear panel of the **REMS** Interface Unit.
- Go to the *Data Collection Setup* tab and set the *TEER Range* selection to Automatic.
- Select the *Take Data/Well Sequence* tab. Press . The electrode travels to the stabilizing well. Press again. The resistance reading (in ohms) of the test resistor displays briefly (Fig. 35). Make a note of the value. The value should be within $\pm 2\%$ of the test resistor value (980–1020 Ω).



Fig. 35 — When you press the Stabilize button, the resistance of the test resistor displays briefly.

- Repeat steps 1 and 2 with the 10 k Ω test resistor. The resistance of the test resistor should be between 9,800 and 10,200 Ω if the unit is functioning properly.
If the resistance displayed on your REMS system falls outside the acceptable limits, please contact technical support at wpi@wpiinc.com or (941) 371-1003.

MAINTENANCE

The REMS should be cleaned occasionally or after accidental spills with a damp, clean cloth and mild detergent.

Replacing a Fuse

A blown fuse may be replaced with a replacement fuse in the fuse holder (Fig. 36). If the replacement fuse blows, it indicates the existence of a serious electrical problem. Call WPI Technical Support at 866.606.1974 or email at technicalsupport@wpiinc.com.

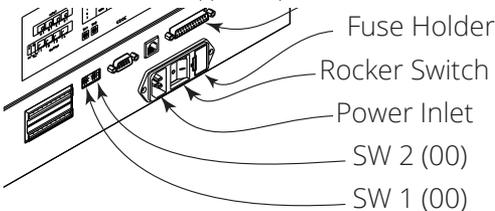


Fig. 36 — Rear panel showing the fuse holder location

- Disconnect the power cord from the power outlet and from the rear panel receptacle.
- Locate the fuse holder on the rear panel (Fig. 36).
- Insert a small screwdriver into the notch located at the left of the fuse holder and twist to remove the fuse holder.
- The fuse holder contains one 2.0A "T" Slo-Blo fuse (5 x 20mm) for a 100/120 voltage selection. It contains two 2.0A fuses for a 220/240 voltage selection.
- Remove the old fuse(s) and insert the new fuse(s).
- Return the fuse holder to its receptacle in the sample changer.

Moving the REMS

If the **REMS** is to be moved, re-install the arm locking screw as follows.

1. Switch off power to the **REMS**.
2. Move the horizontal arm to the right as far as it will go.
3. Locate and remove the arm locking screw from its storage location on the rear panel.
(Shipping Location)

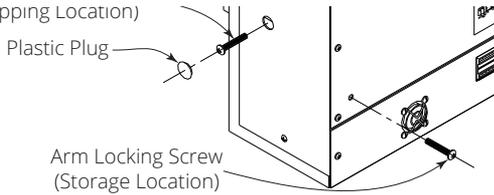


Fig. 37 — The Arm Locking Screw is located on the side panel under a black, plastic plug.

4. Install the arm locking screw into place inside the **REMS** while holding the horizontal arm in its far right position.



CAUTION: When lifting the **REMS**, do not use the horizontal arm as a handle. Grasp the unit by its base.

Packaging for Shipment

NOTE: Use the ORIGINAL packing material for shipping the **REMS** robot.

If the **REMS** is to be packaged for shipment, follow these instructions.

1. Disconnect cable from **REMS** electrode and baseplate.
2. Loosen electrode retaining screw and remove the electrode.
3. Remove baseplate from tray, and then remove the tray.
4. To remove the vertical arm:
 - Remove the plate from the front of the horizontal arm by removing its three screws.
 - Locate the hexagonal-shaped control rod by looking down into the horizontal arm. Lightly press on the rod with your finger and push the rod towards you. When the end of the rod is no longer flush with the end of the horizontal arm, grasp the rod and completely remove it.
 - While holding the vertical arm, loosen the two screws on the underside of the horizontal arm that secures the vertical arm. Remove the vertical arm.
 - Re-insert the control rod as far as it will go. While inserting the rod, you may need to rotate the rod back and forth slightly to get it to pass through the vertical arm's gearing and motor drive socket.
 - Re-attach the plate to the front of the horizontal arm.
5. Carefully package the instrument in its original container.

Electrode Care

Cleaning

After using the electrode, it should be rinsed with distilled water. Over time, the electrode may become coated with media components or proteins that adhere to the surface which can affect performance. To prevent buildup of these materials, we recommend that the electrode be periodically cleaned by soaking the probe tips in an enzymatic detergent like Enzol from Johnson and Johnson or Tergazyme from Alconox. Always rinse the electrode thoroughly with distilled/deionized water as the final step.

Occasionally, the electrodes may be soaked in full strength bleach for 2–5 minutes. This replenishes the chloride layer on the conductive surface of the probes, which helps to maintain charge neutrality. The probes don't need to be exposed to bleach with every cleaning.

1. Rinse the electrode with distilled water or an enzymatic cleaner like Enzol (WPI # **7363-4**).
2. Next soak the electrode in undiluted bleach for five minutes.



CAUTION: Chlorine is corrosive. Avoid contact with metal. Do not let the bleach come in contact with the stainless steel component of the electrode holder.

3. Suspend the electrode in distilled water for a few minutes to remove all the chlorine.

Disinfection

The electrode is non-sterile, as supplied. The electrode **cannot** be sterilized by autoclaving. The electrode may be disinfected using alcohol (one of the most common methods) or a bactericide (for example, Cidex Plus WPI #**7364-4**).

A Typical Disinfection Protocol Using Alcohol



CAUTION: Do not leave the electrode in alcohol for more than 30 minutes. Continuously soaking the electrode in alcohol will weaken the protective coating on the electrode and shorten its lifespan.

1. In a laminar flow hood, immerse the electrode in 70% ethanol for 15 minutes. Allow it to air dry for 15 seconds.
2. Rinse the electrode in a sterile electrolyte solution similar to the experimental cell culture medium or in 0.1M KCl or 0.15M NaCl solution.
3. The electrode is now ready to use.

Storage of the Electrode

The electrode should be rinsed with distilled water and stored dry and in the dark.

ACCESSORIES

Part Number	Description
54320	Replacement baseplate with two rinsing wells
14176	Rinse well cup (polyethylene)
54480	1k Ω Test Resistor
54481	10k Ω Test Resistor
REMS-24	24-Well Electrode and 24-Well Dummy Electrode
REMS-96	96-Well Electrode and 96-Well Dummy Electrode
REMS-96C	96-Well Electrode and 96-Well Dummy Electrode for Corning plates
REMS-CAL	REMS Electrode Calibration Solution

TROUBLESHOOTING

Issue	Possible Cause	Solution
Resistance Measurements Unstable	Electrodes are coated with proteins or other adherent material.	Immerse the probe tips in an enzymatic detergent like Enzo for Johnson and Johnson or Tergazyme from Alconox for several hours. Then, rinse with distilled water and re-test.
	REMS Interface Unit may be faulty.	Use the test resistors to verify that the REMS Interface unit is operating correctly. See "Testing the REMS Electronics" on page 26.
	Electrodes may be worn out or defective.	Run the multi-concentration test to verify electrode performance. Refer to "Multi-Concentration Electrode Performance Testing" on page 24.
Robot Not Responding to Commands	Poor cable connections between computer and robot.	Check that the connectors between the computer and sampler are not loose and that they are fully seated in the sockets.
	Serial communication between the computer and the robot is lost.	Reset the serial communications between the computer and the robot. Check and reset the serial communications status. See "Communication" on page 15.
AutoSampler is dead (Status Light is Out)	The REMS is not getting power.	Verify that the power switch on the back of the REMS AutoSampler is turned on.
		Verify that the AC power cord is securely connected to the REMS and the AC outlet.
		Try a different AC outlet.
		Check the fuse(s). Replace them, if necessary.
Status Light Blinking	An obstruction is hindering free movement.	Check for any obstructions to the horizontal or vertical arms or the electrode.
		Exit the REMS program. Turn off the REMS , wait about 30 seconds, and then turn it on. The status light should stop blinking.
No device listed in NI-DAQ	PCI-6220 board is not installed or is loose	<p>Open the NI-Max program on the desktop. Open the arrow at Devices to see that the NI card is detected and named DEVX. See "Appendix A: Installing the Data Acquisition Card and Software" on page 31. The computer may need to be opened to inspect the card placement.</p> <p>NOTE: Some large Microsoft Windows 10 updates will create a new device "DEV2" in Tab3 (Communications). You have two options. Use that new DEV2. Or you may:</p> <ul style="list-style-type: none"> • Close REMS • Launch Ni-Max to delete DEV1 • Rename DEV2 to DEV1 • Save the NI-Max changes • Relaunch REMS and select DEV1. (Refer to Fig. 38 on page 32 for selecting the PCI 6220 board.)

NOTE: If you have a problem/issue with that falls outside the definitions of this troubleshooting section, contact the WPI Technical Support team at (941) 371-1003 or technicalsupport@wpiinc.com.

SPECIFICATIONS

This unit conforms to the following specifications:

Membrane Resistance Range.....	Auto ranging, or 0 to 2000 Ω and 0 to 20 k Ω
AC square wave current.....	$\pm 20 \mu\text{A}$ @ 12.5 Hz
Electrode positioning.....	Resolution in X, Y and Z: ± 1 mm
Electrode performance.....	Repeatability in X, Y and Z: ± 0.25 mm
Electrode arm speed.....	X- and Y-axis: 250 mm/sec Z-axis: 247.3 mm/sec

Robotic Sampler Technical Data

Control	Computer control via RS-232 port, USB
Manufacturing/ Safety certification:	<ul style="list-style-type: none"> • UL 3101 • CSA 1010 • IEC 1010 - 1
EMC certification:	<ul style="list-style-type: none"> • EN 55011 and EN 50082-1 for CE • FCC part 15 for USA
Dimensions (w x d x h*)	21 ³ / ₃₂ x 17 ³ / ₁₆ x 14 ⁵ / ₈ in. (53.5 x 43.7 x 37.1 cm)
Weight	52 lb. (24 kg)
Line Voltage	User specified, 100/120V or 220/240V
*Height without vertical arm installed	

PC Requirements

- Windows 7, Windows 8 or Windows 10 compatible PC (32 bit/64 bit)
- CPU dual core 3GHz
- RAM 2 GB
- One spare universal or 32-bit PCI slot for the data acquisition card
- RS232 port or USB 2.0 connector (requires a USB2.0- to RS232 convertor)

APPENDIX A: INSTALLING THE DATA ACQUISITION CARD AND SOFTWARE

REMS data acquisition and robot control software is implemented using National Instrument's Labview program. All the National Instruments drivers and utilities for controlling the robot and data acquisition card are included on the REMS software DVD. This process has been performed on delivered REMS systems.

1. Install the software. To do this, simply load the DVD and run the setup.exe program. If it doesn't open automatically, open Windows Explorer and navigate to the DVD drive. Double click on the setup.exe file to run the program.
2. Turn off the computer and unplug it.
3. Install the data acquisition card.
 - Place the computer on an electrostatic discharge (ESD) protected surface that is grounded.
 - Connect yourself to a grounded ESD wrist strap before handling the data acquisition card.
 - Remove the PCI-6220 data acquisition card from its static protective bag.
 - Install the card into any available PCI (not PCIe) card slot.

NOTE: The National Instruments PCI-6220 data acquisition card is universal and will function correctly in 32-bit and 64-bit slots, at either 3.3V or 5V.

4. Reboot the computer. Windows will recognize the presence of the PCI-6220 data acquisition card, and the drivers will be automatically installed.
5. Open the NI-Max program. In the explorer region on the left, navigate to My System>>Devices and Interfaces>>NI PCI-6220 and click on it to open the settings (Fig. 38).

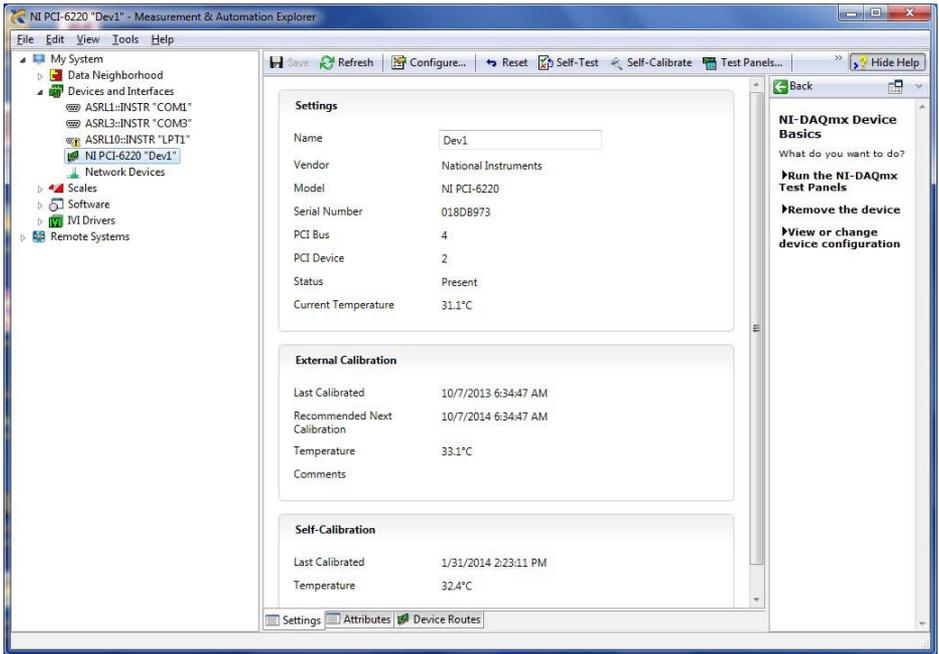


Fig. 38 — In this NI-Max window, the PCI-6220 card is highlighted in the left hand column.

6. In the *Name* field, enter the name of the card so that you will recognize it in the REMS program. In this case, it is named “Dev1.”
7. Click on the *Self-Calibration* button at the top, and allow to card to calibrate. When it completes, a message displays at the top of the window.

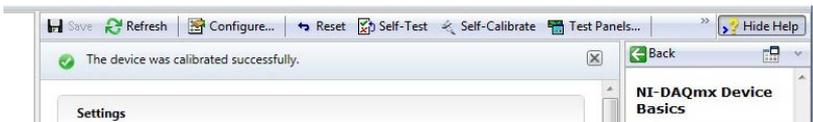


Fig. 39 — A message indicates that the device is calibrated.

8. Press the *Configure* button (at the top of the window) to open the Configuration window. Select the *Power-up States* tab. Choose *port1* from the *Port Name* drop down list. Select the last checkbox (0) in the *Line State* row. This controls the status light on the REMS Interface Unit.

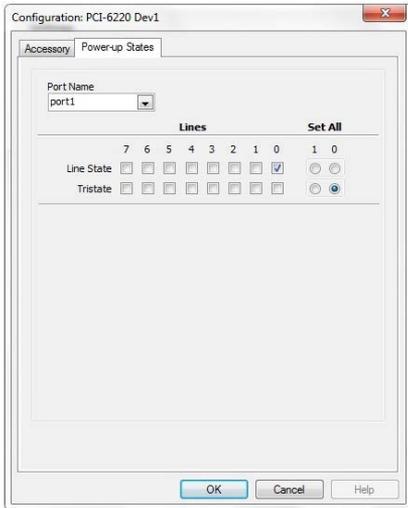


Fig. 40 — Set the last bit in the Line State row.

9. Close the NI-Max software.

APPENDIX B: UPGRADING FROM REMS 6.01 TO 6.02

1. Login as a REMS user with administrator privileges.
2. Uninstall NI serial 4.0. This also removes the existing **REMS** program. If this is not done the following warning appears.

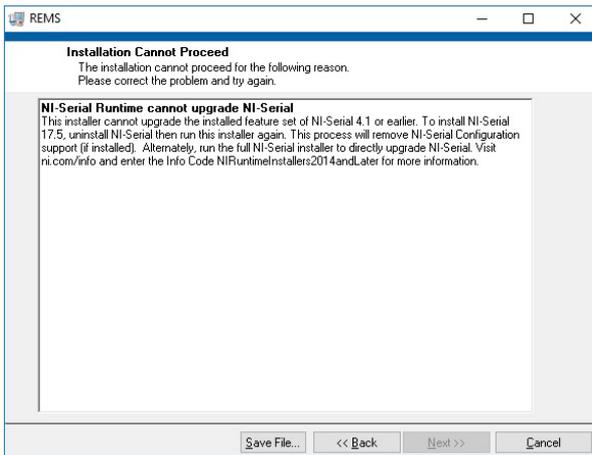


Fig. 41 — A warning appears if you fail to uninstall NI serial 4.0 prior to installing the new REMS version.

NOTE: This uninstall does not affect any prior **REMS** data or plate configuration files. However, it is best to copy the REMS folder (C:\user\REMS\documents\REMS) to your desktop.

To uninstall the NI software, open the National Instruments Software window. Highlight **NI-Serial 4.0** in the Products tab and click on the **X** button to delete the software. The Dependent Products Detail screen appears. Click the **Remove All** button.

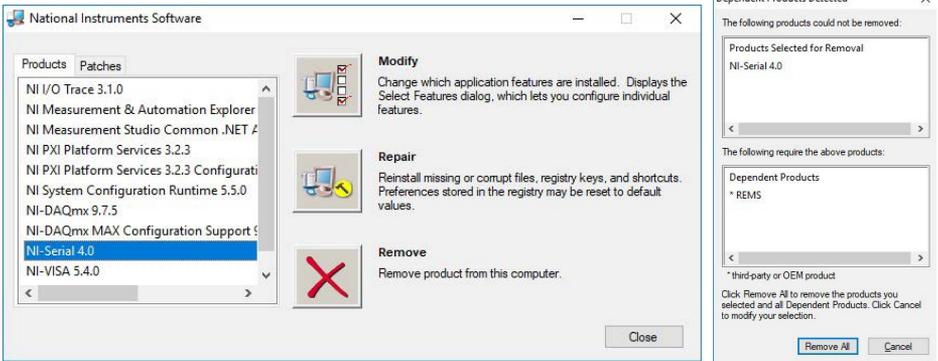


Fig. 42 — (Left) Choose the NI-Serial 4.0 on the National Instruments Software screen and click the X button.
Fig. 43 — (Right) The Dependent Products Detected screen appears.

3. Reboot the PC.
4. Install the new REMS software. The REMS window appears showing the destination directory where the software will be installed. Click the **Next** button.

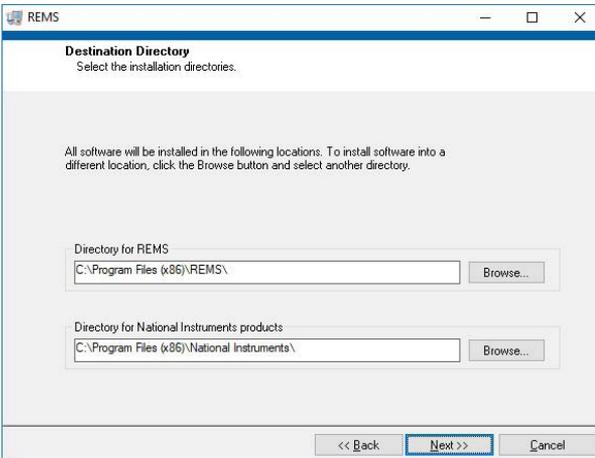


Fig. 44 — The REMS install screen show where the REMS software will be installed.

5. Important information about your system appears on the REMS install screen. Click the **Next** button.

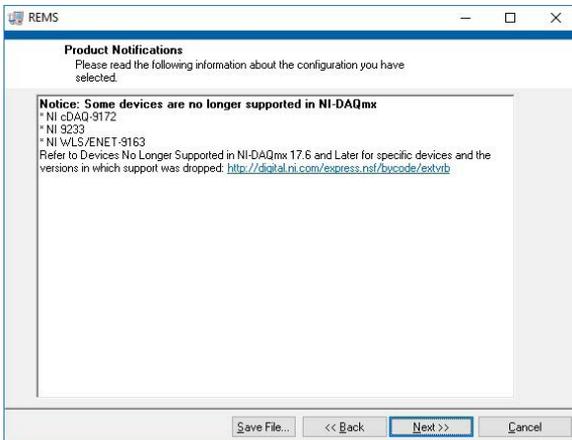


Fig. 45 — The configuration of your system appears on the REMS install screen.

6. The National Instrument License Agreement appears. After reading it, click the acceptance radio button and press the **Next** button.

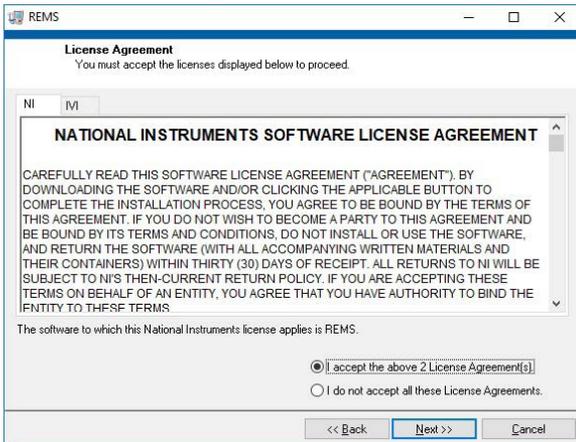


Fig. 46 — The National Instruments License Agreement appears on the REMS install screen.

7. Review the final installation summary and click the **Next** button.

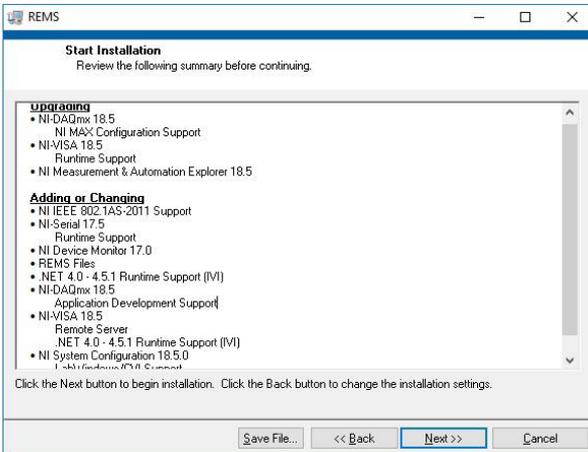


Fig. 47 — An installation summary appears on the install screen.

8. Reboot the PC. After rebooting, launch NI-Max. Then do the following:
- Select **Devices and Interfaces**.
 - Click on the active NI PCI 6220 board and verify that the NI PCI 6220 is named DEV1.
 - If you see DEV2, then delete the inactive DEV1. Then rename DEV2 to DEV1.
 - Save the configuration by clicking the **Save** (disk) button.
 - Push the **Reset** button.
 - Perform a self-test by clicking the **Self-Test** button.
 - Calibrate the software by pressing the **Self-Calibrate** button.
 - Close the program.

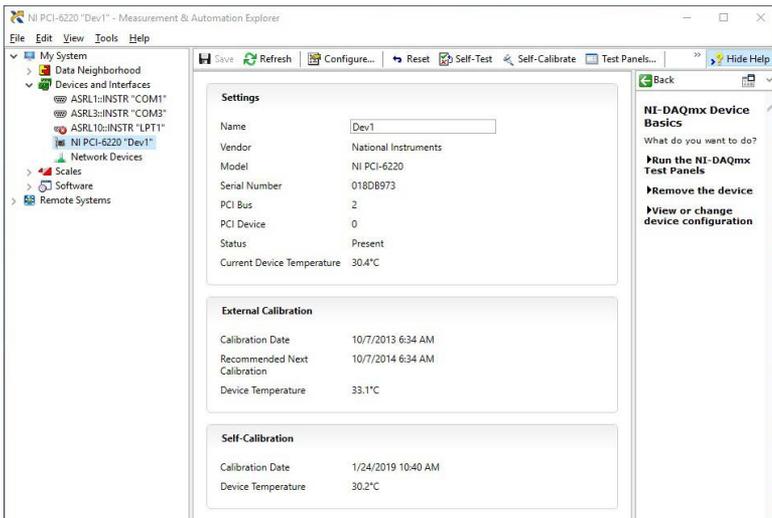


Fig. 48 — The NI Max interface.

9. As an administrator, copy the files in the plate configuration folder located at C:\user\REMS\documents\REMS\Plate_Configurations\ to C:\Users\Public\Documents\REMS\Plate_Configurations. The installation is now complete.

APPENDIX C: ADJUSTING PARALLELISM OF THE WELL PLATE HOLDER

REMS systems are shipped with the parallelism configured at the factory.

The parallelism between the well plate holder and the horizontal axes of the robot is pre-adjusted at WPI. However, a description of the adjustment process is provided here in case you need to perform it. The large plastic tray on the robot is not necessarily perfectly parallel with the robot's horizontal axes. Parallelism of the culture plate relative to the horizontal axes of the robot is critical in order to maintain a consistent probe insertion depth (Z dimension) across the plate. Variations in the Z dimension even as little as 1 mm are undesirable, because they can introduce resistance error into the TEER reading. Adjustments should be made to bring the parallelism between the plate and horizontal arm to a difference of less than 1 mm in all four corners of the well plate holder.

Parallelism between the horizontal arm and well plate holder can be achieved by adjustment of the height of the well plate holder in each corner using (4) height adjustment screws.

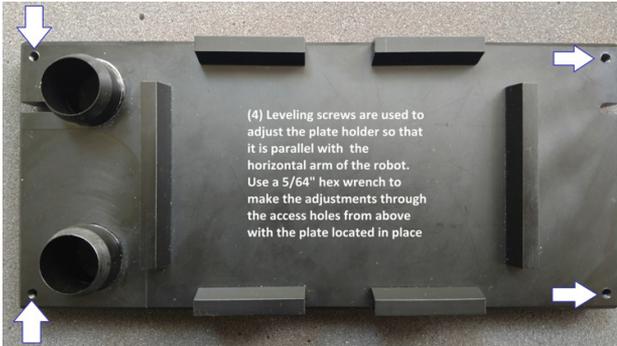


Fig. 49 — Leveling screws are located on the four corners of the well plate holder.

Measuring to Level the Well Plate

Method 1: Locate the horizontal arm over the left and right edges of the well plate holder, and measure the distance of the robot arm down to the upper surface of the well plate holder at the front and rear corners.

A precision ruler may be used. The robot's horizontal arm can be physically positioned at the desired location by manually moving it (with the power off) to the left and right edges of the well plate holder by hand. Take measurements on the front and rear corners on both left and right sides.

Method 2: Alternatively, the robot may be powered up and placed into manual mode to command the robot to move a dummy electrode down to the surface of each of the four corners. This can be done by creating a custom plate configuration file. In a plate configuration file, set the "Z low" vertical axis to position the dummy electrode down close to, but not touching the plate surface. (See "Setting Position Parameters for a New Plate" on page 21.) You may choose to use a shim, like a thick piece of plastic or cardboard (1 or 2 mm thick) or a coin as a spacer between the well plate holder and the electrode. Use the adjustment screws to raise the plate (with spacer in place) up to meet the electrode surface. Repeat this procedure in each of the four corners.

TIP: How to use the robot in manual mode: Under the *Data Collection* tab, place the robot into manual mode by selecting the *Manual* option in the *Sampling Control* drop down menu as described on page 14 of this manual. Then, go to the *Take Data/Well Sequence* tab and click on the *START* button. From here, click on the well that you wish the dummy electrode to move to on the *Data Array Display Box*. (See Fig. 16 on page 10).

Measuring with a Well Plate Installed

The same adjustments described above for the well plate holder may also be conducted with a well plate installed on the well plate holder. The same principles described above apply. Measure the distance from the surface of the well plate to the arm, or use the robot vertical axis with a dummy electrode and a shim.

Leveling the Well Plate Holder

Regardless of which method is used, measurements should be taken at all four corners of the well plate or well plate holder, and the screws should be adjusted so that all four corners are equidistant from the arm horizontal arm or dummy electrode placed directly above the screw access hole.

When adjustments are complete, the well plate holder must be stable (no rocking). The location of the (4) adjustment screws is shown in Fig. 49 on page 37. If desired, Loctite 222 (WPI# **7004**) may be used to secure the adjustment (Fig. 50).



Fig. 50 — The leveling screw, secured with Loctite, is shown from the underside of the well plate holder.

APPENDIX D: PROGRAM NOTES FOR IT DEPARTMENTS

User Access Rights

Only the system installer needs to have administrator rights to the PC. All user accounts may have user privileges. The REMS administrator account can now be password protected (as of version 6.02).

Some new antivirus programs lock out programs under ransomware or file protection tools. The NI background programs must not be affected by those antivirus programs, or the **REMS** or NI-Max programs will not operate.

CAUTION: Do not install or reinstall the program for each user.

CAUTION: Do not install the Gilson or the NI disk programs. Doing so could change the robot's home position and negate the plate configuration file or interfere with operation of the REMS program. It is not advised to update the NI program package. Before doing so, contact WPI Technical support. Please send a screen shot of the program and the revision(s) to be updated.



CAUTION: Do not edit or delete the plate configuration files outside of the REMS program.

Universal access to the files (as of version 6.02)

The folder with the plate configuration files is now available in a location that allows universal access to all REMS (version 6.02 and greater) users.

NOTE: REMS users must be able to modify this file to program the XYZ location of the probe's A1 well. This is necessary if you have mechanical changes to the probe or the robot.

The structure of this folder for user REMS is as follows:

C:\users\Public\Documents\REMS\

Plate_Configurations Plate_name_file.txt is calibrated by WPI for the probe type, plate type and robot offsets.

Plate_Data Files contain user-collected plate resistance data.

Plate_Background Files contain user-collected plate background resistance data.

Files stored in each users Documents directory

If you prefer, the folder with the plate configuration files may be directed to each users Documents directory for data storage. Then, these files are copied when a user launches the REMS program.

NOTE: These path dialog box entries show the folder path names only and do not contain file names.

1. To change that path correctly, select the browse button next to the Data Folder field on the Data Storage tab (Fig. 51).

D	C	B	A	
4	3	2	1	1
8	7	6	5	2
12	11	10	9	3
16	15	14	13	4
20	19	18	17	5
24	23	22	21	6

Fig. 51 — The Data Storage tab shows the folder where plate data is stored. The arrow indicates the Browse button.

2. Navigate to the new location for the file storage location. Select the folder from the left panel. Then, click the Current Folder button.

If you have questions about this procedure, please contact technical support at technicalsupport@wpiinc.com.



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DECLARATION OF CONFORMITY



WORLD PRECISION INSTRUMENTS, INC.

175 Sarasota Center Boulevard
Sarasota, FL 34240-9258 USA
Telephone: (941) 371-1003 Fax: (941) 377-5428
e-mail wpi@wpiinc.com

DECLARATION OF CONFORMITY

We: World Precision Instruments, Inc.
175 Sarasota Center Boulevard
Sarasota FL 34240-9258
USA

as the manufacturers of the apparatus listed, declare under sole responsibility that the product(s):

**Title: SYS-REMS Automatic Tissue Resistance Measuring System,
consisting of the following component parts:**

- 1. Part No. 54006 REMS Robotic Sampler Assembly**
- 2. Part No. 54320 Tray Holder Assembly**
- 3. Part No. 54380 REMS Electronic Unit Assembly**
- 4. Associated interunit cabling**

to which this declaration relates is/are in conformity with the following standards or other normative documents:

Safety:	EN 61010-1:2001
EMC:	EN 61326:1998/A2:2001/A3:2003
	EN 55011:1998/A1:1999/A2:2002
	EN 61000-6-1:2001
	EN 61000-6-3:2001

and therefore conform(s) with the protection requirements of Council Directive 2004/108/EC relating to electromagnetic compatibility and Council Directive 73/23/EEC relating to safety requirements.

Issued on: May 16, 2006

Mr. Harry Fein
President

World Precision Instruments, Inc.
175 Sarasota Center Boulevard
Sarasota, FL 34240-9258 USA

Mr. Glen Carlquist
Production Manager

World Precision Instruments, Inc.
175 Sarasota Center Boulevard
Sarasota, FL 34240-9258 USA

WARRANTY

WPI (World Precision Instruments, Inc.) warrants to the original purchaser that this equipment, including its components and parts, shall be free from defects in material and workmanship for a period of one year* from the date of receipt. WPI's obligation under this warranty shall be limited to repair or replacement, at WPI's option, of the equipment or defective components or parts upon receipt thereof f.o.b. WPI, Sarasota, Florida U.S.A. Return of a repaired instrument shall be f.o.b. Sarasota.

The above warranty is contingent upon normal usage and does not cover products which have been modified without WPI's approval or which have been subjected to unusual physical or electrical stress or on which the original identification marks have been removed or altered. The above warranty will not apply if adjustment, repair or parts replacement is required because of accident, neglect, misuse, failure of electric power, air conditioning, humidity control, or causes other than normal and ordinary usage.

To the extent that any of its equipment is furnished by a manufacturer other than WPI, the foregoing warranty shall be applicable only to the extent of the warranty furnished by such other manufacturer. This warranty will not apply to appearance terms, such as knobs, handles, dials or the like.

WPI makes no warranty of any kind, express or implied or statutory, including without limitation any warranties of merchantability and/or fitness for a particular purpose. WPI shall not be liable for any damages, whether direct, indirect, special or consequential arising from a failure of this product to operate in the manner desired by the user. WPI shall not be liable for any damage to data or property that may be caused directly or indirectly by use of this product.

Claims and Returns

Inspect all shipments upon receipt. Missing cartons or obvious damage to cartons should be noted on the delivery receipt before signing. Concealed loss or damage should be reported at once to the carrier and an inspection requested. All claims for shortage or damage must be made within ten (10) days after receipt of shipment. Claims for lost shipments must be made within thirty (30) days of receipt of invoice or other notification of shipment. Please save damaged or pilfered cartons until claim is settled. In some instances, photographic documentation may be required. Some items are time-sensitive; WPI assumes no extended warranty or any liability for use beyond the date specified on the container

Do not return any goods to us without obtaining prior approval and instructions from our Returns Department. Goods returned (unauthorized) by collect freight may be refused. Goods accepted for restocking will be exchanged or credited to your WPI account. Goods returned which were ordered by customers in error are subject to a 25% restocking charge. Equipment which was built as a special order cannot be returned.

Repairs

Contact our Customer Service Department for assistance in the repair of apparatus. Do not return goods until instructions have been received. Returned items must be securely packed to prevent further damage in transit. The Customer is responsible for paying shipping expenses, including adequate insurance on all items returned for repairs. Identification of the item(s) by model number, name, as well as complete description of the difficulties experienced should be written on the repair purchase order and on a tag attached to the item.

* *Electrodes, batteries and other consumable parts are warranted for 30 days only from the date on which the customer receives these items.*

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