

INSTRUCTION MANUAL

EVM-MT-03-02

EVOM[™] Manual for TEER Measurement with Secure Data Transfer Capability

> www.wpiinc.com 012825

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QUICK START FOR TAKING MEASUREMENT

- 1. Power on the **EVOM™ Manual** using the power switch on the rear panel.
- 2. To configure your data file storage settings, hold the *Store* button on the main screen for 2 seconds.
 - Select the preferred storage mode, *USB Save* or *APP Save*. See "Storing Data" on page 15.
 - If a filename other than the default (plate1) is required, choose whether you
 will use a prefix and auto-indexing or a unique file name. To use a prefix and
 auto-indexing, activate the *Auto Index* radio button. Then, press *Store Prefix* to
 enter a file name. To use a unique file name, deactivate the *Auto Index* radio
 button. Then, select *File Name* to enter a new name.
- 3. Prepare the storage location you selected above, either USB or PC using the appropriate method:
 - If you selected USB Save, you need to insert the USB flash drive into the USB port on the side of the EVOM™ Manual. See "Storing Data on the USB Flash Drive" on page 17.
 - If you selected *APP Save*, need to make sure the **EVOM™ Manual** is properly connected to the computer using the USB cable included and verify that the Companion Application is running. See "Storing Data on a Connected PC" on page 19.
- 4. Configure your plate settings, including the number of wells, the mode (resistance or voltage), the mode units, and blank handling, and perform any electrode preparation.
 - For taking resistance measurements, refer to "Resistance Configuration Procedure" on page 27
 - For taking voltage measurements refer to "Voltage Configuration Procedure" on page 28.
- 5. Whether you are taking resistance or voltage measurements, the data acquisition procedure is identical. To begin measuring with well A1, do one of the following:
 - Press the foot switch or touch the *Store* button on the main screen. A
 measurement of the first well is taken, and the data storage advances to the
 next well in your sequence. Tap the *Store* button again to measure the next
 well in your defined sequence.

NOTE: When required, you may use the *Previous Well* area on the left side of the screen to return to the last well and re-measure or the *Next Well* area on the right side of the screen to skip a well. Then press the *Store* button or the foot switch to take the measurement of that cell.

 Press the Preview button to see a graphical display of the plate. The green cell indicates the cell to be sampled, and the yellow cell indicates the next well in the sequence. Press the foot switch to take a measurement and advance to the next well. Repeat the sampling process. Press the *Preview* button to see the measurements that have been taken.

- 6. Once the last well has been recorded, the file storing selection notification appears prompting you to take an action. Select one of the options as follows:
 - *Export* starts the storage of all the plate files. When you select it a message appears describing which plate is being stored. The number of plates could be one, two or three, depending on how many were collected before exporting.
 - *Next* returns you to the main screen so you can take measurements of the next plate which starts from a clear state.

NOTE: If you are on plate 3, the *Next* button does not appear, because EVOM™ Manual's memory only holds data from three plates.

- **Back** returns you to the main screen (data collection). You go back to the first well of the current plate being collected and can acquire the data again.
- 7. When the plate data has been saved, a confirmation message appears. Press the *Done* button to return to the main screen.
- 8. Now you can work with your data.
 - If you stored your data on the USB flash drive, remove the drive and transfer the data files to your computer. They are in comma separated value (CSV) file format for Microsoft[®] Excel.
 - If you stored your data using the EVOM[™] Companion Application, all your data files will be located in the directory you specified when you started the Companion Application.

TEER Calculations

To compute TEER, multiply the measured resistance by the surface area's listed below. For example, a 12 mm insert measures 565 Ω , the TEER is 565 Ω * 1.13 or 638 Ω -cm².

- 6 well plate (24 mm inserts) 4.52 cm²
- 12 well plate (12 mm inserts) 1.13 cm²
- 24 well plate 6.5 mm inserts) 0.3316 cm²
- 96 well plate (4.3 mm inserts) 0.145 cm²

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 or data. Please read these carefully.

 \triangle

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.



Fig. 1—EVOM™ Manual with a STX4 electrode.

INTRODUCTION

EVOM™ Manual is the WPI's manual instrument for measuring Trans Epithelial Electrical Resistance (TEER). The **EVOM™ Manual** has been engineered to facilitate ease of accurate, repeatable measurements and transfer of data. You no longer need to log data by hand. The resistance or voltage information is written to a USB drive in a format (CSV) that can be opened by most spreadsheet programs on either PC or Mac. The processor controlled instrument is more stable and accurate. Included is a footswitch for quick hands-free recording to the USB flash drive. The touch screen interface is easy to use and setup for setting control wells in resistance or voltage modes and measurements. The auto ranging resistance feature allows for fast resistance measurements, and an overrange display feature ensures no false readings. The **EVOM™ Manual** now has adjustable current levels in three fixed ranges with two lower ranges for sensitive membranes and high resistance ranges up to 100KΩ.

Confluence of a cellular monolayer is determined by an increase or a plateau in tissue resistance detected using the unique electronic circuit of the **EVOM™ Manual** and the **EVOM™ STX4** electrode. The **EVOM™ Manual** qualitatively measures cell monolayer health and quantitatively measures cell confluence. The **EVOM™ Manual** produces a low AC current that avoids electrode metal deposits and adverse effects on tissues which can otherwise be caused by higher DC currents. The **EVOM™ Manual** uses low current and voltages and is designed for non-destructive testing for epithelial monolayer confluence in cell cultures. In addition, resistance readings are unaffected by membrane capacitance or membrane voltage. The accuracy and repeatability of the **EVOM™ Manual** system makes this instrument ideal for permeability, potential difference (PD) and other detailed membrane studies.

EVOM™ Manual Features

- Low noise design offers greater resolution and accuracy
- · Automatic 20X sample averaging improves accuracy and stability
- Adjustable fixed measurement currents (2, 4 or 10 μA)
- Resistance auto ranging from 1 Ω to 100,000 Ω or with three fixed current ranges
- Reliable low current, low voltage design prevents metal ion transport
- Fast resistance stabilization on low levels under 200 Ω with resolution to 0.1 Ω
- · Ergonomic tilt stand for low glare operation
- Graphical display of popular plates (6, 12, 24, 96) for trend analysis
- Display shows the most recent set of parameters
- Automatic plate indexing operation with or without control well subtraction for resistance and potential difference (PD) measurements
- Continuous data logging via USB (PC, Mac, Linux)
- · Saves date stamped data to a spreadsheet readable file on a USB drive
- Upgradable firmware
- Secure data transfer to PC via the EVOM™ Companion Application

EVOM™ Manual Benefits



Eliminates errors and reduces experimental processing time



Auto data logging eliminates the need to track data by hand



The small footprint allows more bench space



Easy calibration and verification



Footswitch for hands-free recording



Prevent data loss with auto save and data recovery when battery is low

TEER is easily computed by applying a unit area formula to the resistance

Notes and Warnings

Choice of current/range: The EVOM™ Manual has three ranges of current from

2 μ A to 10 μ A and each current range has its own resistive upper limit up to 100K Ω :

- 10K Ω range = 10 μA
- 50K Ω range = 4 μ A
- 100K Ω range = 2 µA

The auto range will adjusts the current as needed from 2 μA to 10 μA depending on the resistance.

General rules

- The higher the current, the greater the stability of the measurement. The fixed ranges may be more consistent with a fixed current for some cell lines.
 - If a cell line is not expected to read above 10K Ω , then choose that fixed range.

Use of the charger: The **EVOM™ Manual**'s Li-ion battery lasts 8 hours on its own. The charging time is 5.5 hours with the unit powered off. The **EVOM™ Manual** charger should not be plugged in all the time. The charger may be used to power the **EVOM™ Manual** for normal and long-term use.

NOTE: In the higher resistance ranges (50K -100K) there may be a slight instability added from the noise of the power supply, in that case unplug it during those measurements.

USB Drives: WPI has supplied a tested USB flash drive. Most drives of 32 GB (USB2.0) or smaller sizes have been tested.

CAUTION: Not all USB drives work on the EVOM™ Manual.

To test a USB drive, see "Sample Resistance File (*.csv)" on page 23. This test saves a file to the drive and reads it back. A system message reports if the USB flash drive is usable, or it will display an error. If the USB flash drive is not plugged in or it is not recognized, a warning message will appear. Press *Cancel* and try again. The *Store* function cannot be used on an unrecognized drive. **EVOM™ Manual** should respond with "File saved" within 15 seconds. If "Saving file..." is still seen after 20 seconds, press the *Return* key and use another USB flash drive. Once a file has been saved, open the file on a computer to verify the contents.

Tips and Advice

Out of Range Error

If you see ---- (5 dashes) on the main resistance display, then the resistance reading is out of range. Check the ohms range setting to see if it can be increased to display a higher resistance or set it to *Auto*. If the display still shows ----, then go to the *Blank Handling* screen and check the settings and the reading. A saved reading will show a space ("") in the grid if the display shows out of range. Do not forget to reset the blank to zero or go back to a control well resistance when you are finished.

NOTE: The 100K Range can read 5% over to 105K Ω .

Zeroing the blank handling value

- 1. Select Setup.
- 2. Then, press *Short Electrodes*.
- 3. Press the *Blank Handling* button.
- 4. Set the reading to zero.
- 5. Return to the *Setup* page and open the electrodes to remove the short.

Zeroing the probe null offset

- 1. Select Setup.
- 2. Then, press *Short Electrodes*.
- 3. Press the *Probe Null* button.
- 4. Set the reading to zero.
- 5. Return to the *Setup* page and open the electrodes to remove the short.

Resistance levels that are unexpectedly lower or higher can be attributed to a number of things besides an incompletely grown membrane. Any of these items can change the resistance reading:

- Electrode resistance (Keep the electrodes clean.)
- Fluid levels (Try to maintain the same volume for every insert. A variation of 50 μL can have a large effect in a small insert.)

- Changing CO₂ levels and temperature drift can account for large changes in the resistance reading. The use of a warming plate can reduce this effect.
- We recommend that both fluid levels are at the same height so that you do not have any pressure differentials and the apical well is filled first so that you do not dislodge the membrane from the filter by hydrostatic pressures. If the membrane detaches itself, then it was not adhering to the inserts filter. A small gap from dislodged tissue at the edge of the membrane can open a clear path for the EVOM™ Manual to read a lower resistance. Instead of the current going through the membrane, the electricity is going around the membrane. Electricity will take the path of least resistance and the EVOM™ Manual will measure that as a lower resistance.

NOTE: Cleaning the electrode often corrects most of these issues. Refer to the Maintenance section of the electrode manual for cleaning protocols.

Charging the EVOM™ Manual

The **EVOM[™] Manual** battery may also be charged with a Mini-B USB cable (WPI #**803026**), not supplied.

Parts List

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

(1) EVOM™ Manual Epithelial Volt Ohm Meter

(1) **97893** Programmed USB drive 32 GB (For USB data storage, and contains the EVOM[™] Companion Application and a Python 3.8 program for continuous digital monitoring of a target insert. See "Appendix B: Continuous Recording via USB (Python) on a PC" on page 42.)

- (1) **503535** USB cable
- (1) **99673** 1000Ω Test Resistor
- (1) 803025 A/C power cord and charger
- (1) **13142** Foot switch

Instruction Manuals for the **EVOM™ Manual** and **STX4** electrode can be downloaded at https://www.wpiinc.com/manuals.



Fig. 2—The EVOM™ Manual includes these parts.

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 51 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 100 mm (4") of shock absorbing material. For further details, please read the section entitled "Claims and Returns" on page 51 of this manual.

INSTRUMENT DESCRIPTION

EVOM[™] Manual Meter

The EVOM[™] Manual meter is compact and portable, and setup is quick.



Fig. 3—The ports on the side of the unit let you connect a USB flash drive and the EVOM™ Electrode.

The side panel connection ports include:

- USB Port for Flash Drive Insert the USB flash drive (WPI #97893) into this port when you wish to collect data on the flash drive. Make sure it is properly seated.
 NOTE: The USB flash drive contains the files for installing the EVOM™ Companion Application that is used for secure data transfer to a connected PC. When you need to install those files on a PC, plug the USB flash drive into any USB type A port on the computer to access the files. See "Appendix F: Installing the EVOM™ Companion Application" on page 49.
- **Electrode Port** Plug the STX series electrode or EndOhm chamber into this port when you are taking measurements. You may also plug the 1000Ω test resistor into this port when you are calibrating the unit.



Fig. 4—The ports and controls on the back of the unit let you connect a foot switch, connect a USB cable to a PC for secure data transfer, connect the power adapter, and power on the system.

The back panel has the following connection ports and switches:

- Foot Switch Port Plug the foot switch (WPI #13142) into this port for a simple, hands-free way to take measurements.
- USB Port for Secure Data Transfer to PC This USB type B port is used for secure data transfer. Use the included USB cable (WPI #503535) to connect the EVOM™ Manual to your PC. The type A end plugs into a port on the computer, and the type B end plugs into this port on the EVOM™ Manual.
- **Programming Button** FOR FACTORY USE ONLY. Do NOT push this button without having specific instructions from WPI.
- **Power Connection Port** Plug the A/C power cord (WPI **#803025**) into this port to charge your **EVOM™ Manual**.

CAUTION: Always connect power when the warning messages appear. NEVER let the battery get completely discharged, or it may not charge again. See "Power Level Messages" on page 32.

• **Power Switch** – Toggle this switch to turn the unit off (O) or on (|).

Theory of Operation

The **EVOM™ Manual** system uses an electrode assembly made of four subelectrodes, two for current sourcing and two for voltage measurement.

The **EVOM™ Manual** passes a known constant current through the membrane on two electrodes and measures the voltage needed to pass that current on the other two electrodes and computes resistance using Ohm's law. The polarity of the current changes from positive current to negative current 12.5 times per second. This polarity change avoids leaving a charge on the membrane and negates any voltage offsets due to the membrane potential or from the electrodes. The amount of current and voltage is low (2, 4 or 10 µA) to minimize any tissue stimulation and to prevent the migration of metal ions.

SYSTEM SETUP-

Main Display



Fig. 5—The main EVOM™ Manual display.

A negative resistance number will appear in red text. This can be caused by a faulty electrode in media, (see "System Messages" on page 32) or a blank setting that is over the measurement (see "Blank Handling Page" on page 14). When the meter is plugged into a power source, the battery icon shows a charging symbol.

Setup Page

Press Setup on the main screen to open the Setup page.



Fig. 6—Setup page

- Select Plate Select the number of wells and the sampling pattern.
- Mode Units Choose the Units for sampling. (Options include Millivolts, Volts, Ohms and K Ohms.)
- **Blank Handling** Lets you subtract the control blank from the current resistance measurement.

- Short Electrodes Shorts V_1-V_2 and is used to nullify the galvanic charge in the electrodes when the electrodes are placed in a saline solution for 4–8 hours to remove the charge. See also "Probe Null" on page 15.
- Date/Time Select the Date and Time formats for your data files.
- **Calibrate** Calibrate the electrode. (This uses a 10K Ω 0.1% resistor.) See "Instrument Diagnostics and Calibration" on page 25.
- Probe Null Neutralize the offset charge on V₁-V₂.
- **Store Screen** Store data files, configure how the files are named and if they are automatically numbered.

Plate Selection Page and Plate Preview

1. Press Select Plate on the Setup page to access the Plate Selection screen.



Fig. 7—*Choose the number of wells and the sampling pattern.*

- 2. Select the radio button for the number of wells in your plate.
- 3. Select the sampling pattern from the options in the bottom half of the window.
- 4. Press *Return* to save the setup. When you press *Preview* from the main screen you will see the plate preview files like the samples below.



Fig. 8—(Left) 6-well plate preview Fig. 9—(Right) 12-well plate preview

3775		1	2	3	4	5	6		_	1	2	3	4	5	6
		0	0	0	0	0	0		•	0	0	0	0	0	0
				."					в	0	0	0	0	0	0
z	в	0	0	0	0	0	0	Z	с	0	0	0	0	0	0
~	c	0	0	0	0	0	0	۲,	D	0	0	0	0	0	0
2		-		-		-	-	Ē	E	0	0	0	0	0	0
Ξ.	D	U	U	U	U	U	U	ш	F	0	0	0	0	0	0
~								-	G	0	0	. 0	0	0	0
			A	1=1	1000)			н	0	0	0	0	0	0
											1	41 =	1000)	

Fig. 10—(Left) 24-well plate preview *Fig.* 11—(Right) 48-well plate preview

		1	2	3	4	5	6	
	A	0	0	0	0	0	0	
	в	0	0	0	0	0	0	
Z	С	0	0	0	0	0	0	
4	D	0	0	0	0	0	0	
F	E	0	0	0	0	0	0	
	F	0	0	0	0	0	0	
	G	0	0	0	0	0	0	
	н	0	0	0	0	0	0	
		< Scrol		41 =	1000		Scroll >	

Fig. 12—96-well plate preview. (Press scroll to see the additional wells.)

NOTE: An out of resistance range displays ---- (5 dashes) on the screen. Adjust the range to correct the error. A saved well under preview will show a space (" ") if the display was recorded out of range. The 100K range can read 5% over the 105K Ω .

Mode Units

1. Press *Mode Units* on the *Setup* page to access the *Select Volts or Ohms* screen.



- Fig. 13—Choose the unit of measurement and (if you are using Ohms) the range.
- 2. Choose your units. Options include Millivolts, Volts, Ohms and K Ohms. If you choose ohms, then you must also select an Ohms Range.

Fiz	xed Current	Va	riable Current
10 K	10 µA	Auto	2 µA to 10 µA
50 K	4 µA		
100 K	2 IIA		

3. Press *Return*.

Blank Handling Page

1. Press *Blank Handling* on the *Setup* page to subtract the control well blank from the current resistance measurement.





- Fig. 14—(Left) Blank Handling window.
- Fig. 15—(Right) The main screen shows the offset underneath the reading.
- 2. Insert the probe into an empty well filled with media. Press *Reading* to make a measurement.
- 3. Select the *Blank Enable* radio button to enable the offset.
- Press *Return*. The resistance offset shows under the reading on the main screen. If the blank (offset) is greater than the resistance reading, the reading displays negative numbers in red text. The saved data file reflects the subtraction. NOTE: This includes fluid and electrode resistances.

Setting the Date and Time

1. Press *Time/Date* on the *Setup* page to set the date and time format.



- Fig. 16—The Set Date/Time screen lets you set the date and time and choose the display format.
- 2. Press *Set Date* (Fig. 17). Set the appropriate date and press *Return*.
- 3. Press Date Format (Fig. 18). Choose your preferred format and press Return.



Fig. 17—(Left) Set the date and press Return Fig. 18—(Right) Choose the date format and press Return

- 4. Press *Set Time* (Fig. 19). Set the appropriate time and press *Return*.
- 5. Press *Time Format* (Fig. 20). Choose 12 hour or 24 hour and press *Return*.





Fig. 19—(Left) Set the time and press Return Fig. 20—(Right) Choose the time format and press Return

Probe Null

1. Press Probe Null on the Setup page to access the NULL Probe Process screen (Fig. 21).



- *Fig. 21—(Left) Null probe process.*
- Fig. 22—(Right) The potential difference offset shows on the main screen under the reading.
- 2. Place the electrodes in a saline media.
- 3. Push the *NULL* button. Wait for the second message stating "Solution value 0.0" before returning. This action removes any voltage offsets on the electrodes and neutralize the offset charge on V_1 - V_2
- 4. Select the *Enable/Disable* radio button to use the offset.
- 5. Press *Return*. The potential difference offset shows on the main screen under the reading. An electrode check should show 0.0 mV. The saved data file reflects the offset (Fig. 22).

Storing Data

The **EVOM™ Manual** has two options to save data.

- Data may be saved to a USB flash drive plugged into the USB port on the side of the unit.
- Data may also be stored on a PC with the use of the EVOM™ Companion Application. The EVOM™ Manual can store up to three plates of data without

having to export or transfer the data to an external storage device. Each plate is saved as an individual .CSV file which may be opened in Microsoft[®] Excel or another spreadsheet application.

1. Press and hold the *Store* button on the main screen for 2 seconds or press *Store Screen* on the *Setup* screen to configure your stored data files.



- Fig. 23—Use the Store Options screen to save a data file.
- 2. Press *New Plate* to clear any recorded data on the preview screen and from memory.



- 3. Select either the *USB save* or the *APP save* radio button.
 - **USB save** stores the files to the USB flash drive. Make sure the USB flash drive is installed.
 - APP save stores the files on a connected PC using the EVOM™ Companion Application. Be sure to properly connect the EVOM™ Manual to the PC and start the Companion Application before storing measurements.
- Press *File Name* to enter a unique name for your data file. In the entry page, select *Clear*, then type the file name, and press *Enter* when you finish. If you choose to use your file name, auto-indexing must be disabled.



Fig. 24—Press Clear. Type the desire name. Press Enter.

5. If you prefer to use the *Auto Index* function, press *Store Prefix* to enter a unique file name that will be appended with sequential numbers each time your file is saved. Select the *Auto Index* radio button to enable the *Store Prefix* and *Auto Index* buttons.

NOTE: When the *APP save* mode is selected, *Auto Index* is automatically selected. It is the only file name method used when saving data files to a connected PC.

- 6. If you are using the USB save mode, you may select the *Auto Index* radio button if you want the **EVOM™ Manual** to automatically number the new data files sequentially. Auto indexing changes the name to a numeric sequence formatted with a prefix and a sequential number (prefixN).
- 7. Press the *Return* button to go back to the main screen or press the *Store Now* button to save the current data set. The prompt you see depends on the storage mode you chose in step 3.
 - For USB Save, see "Storing Data on the USB Flash Drive" on page 17.
 - For APP Save, see "Storing Data on a Connected PC" on page 19.

NOTE: Since there are three plates that can be held in the EVOM[™] Manual's memory before data is exported to a storage device, there could be as many files as the number of plates analyzed kept in the memory. A separate file is generated for each plate in memory, each with a distinct file name.

Storing Data on the USB Flash Drive

1. If you selected the *USB Save* option (Store Options screen) and pressed the *Store* button on the main screen or the *Store Now* button on the Store Options screen, a prompt appears asking you to insert the USB flash drive (Fig. 25).



Fig. 25—Verify that the USB drive is inserted correctly into the USB port on the side of the EVOM™ Manual.

NOTE: When you are using standard file naming instead of auto-indexing, a suffix of -1, -2, or -3 is appended to the file name to indicate which of the three stored plates corresponds to each file.

NOTE: If you try to save a second set of plates with the same filename, a warning screen displays indicating that a file with the same name already exists (Fig. 26). You may press the *Back* button to return to the main screen. Then, hold the *Setup* button for 2 seconds to access the *Store Options* screen and change the name. When you store the files, you are asked to confirm the write operation for each file to be written separately.

File Exists,	Overwrite?
EXPORT	ВАСК

Fig. 26—To overwrite the existing file with the same name, press the Export button. Otherwise, press the Back button, return to the Store Options screen and change the name.

- 2. Select one of the options as follows:
 - *Export* starts the storage of all the plate files, overwriting any duplicate files. When you select it, a message appears describing which plate is being stored (Fig. 27). The number of plates could be one, two or three, depending on how many were collected before exporting.
 - Next returns you to the main screen so you can take measurements of the next plate which starts from a clear state. The note above the Setup button on the main screen indicates which plate data is being collected (Fig. 5).

NOTE: If you are on plate 3, the *Next* button does not appear, because **EVOM™** Manual's memory only holds data from three plates.

• **Back** returns you to the main screen (Data Collection). You go back to the first well of the current plate being collected and can acquire data.



Fig. 27—When you press Export, the message indicates which plate data is being stored.

NOTE: If the USB fails to write the data in the set time out period, the "Saving file..." notice will not change to "File saved." A file is typically saved in less than 20 seconds. If your file does not save, press the *Back* button and use a different USB flash drive.

3. Once the data is transferred to the USB flash drive, the message displays to indicate the files were saved successfully (Fig. 28).



Fig. 28—This message indicates the plate(s) was/were saved to the USB.

Storing Data on a Connected PC

Before storing EVOM[™] data files on a connected computer, the **EVOM[™] Manual** must be connected to the computer and the EVOM[™] Companion Application must be running on the computer. See "Starting the EVOM[™] Companion Application" on page 20.

 If you selected the *APP Save* option (Store Options screen) and pressed the *Store* button on the main screen or the *Store Now* button on the Store Options screen, a prompt appears asking you to connect to the Companion Application (Fig. 29).



Fig. 29—Verify that the EVOM™ Manual is connected to the EVOM™ Companion Application running on the computer.

- 2. Select one of the options as follows:
 - *Export* starts the storage of all the plate files. When you select it, a message appears describing which plate is being stored (Fig. 30). The number of plates could be one, two or three, depending on how many were collected before exporting.
 - Next is available on plate 1 and 2, but not on plate 3. It returns you to the main screen so you can take measurements of the next plate which starts from a clear state. The note above the Setup button on the main screen indicates which plate data is being collected (Fig. 5).

NOTE: If you are on plate 3, the *Next* button does not appear, because **EVOM™ Manual**'s memory only holds data from three plates.

• **Back** returns you to the main screen (Data Collection). You go back to the first well of the current plate being collected and can acquire data.



Fig. 30—(Left) The message changes showing which plate is being saved. Here Plate 3 data is being stored on the connected computer.

Fig. 31—(*Right*) All three plates were saved to the connected computer.

3. When the plate data has been saved, a confirmation message appears (Fig. 31). Press the *Done* button to return to the main screen.

NOTE: EVOM™ Manual has a time out in case the PC is not connected or the EVOM™

Companion Application is not running. If this error occurs, a message appears describing of the communication error (Fig. 32). If this happens, make sure the USB cable is connected to both the **EVOM™ Manual** and the computer, and verify that the application is running. Then press the *Export* button again.



Fig. 32—This message indicates that the data save has timed out. Make sure the unit is connected to the PC and the EVOM™ Companion Application is running on the PC.

Starting the EVOM[™] Companion Application

To be able to save files to a PC, the EVOM[™] Companion Application must be running on the PC. To install the EVOM[™] Companion Application, see "Appendix F: Installing the EVOM[™] Companion Application" on page 49.

1. Launch the EVOM[™] Companion Application by typing "EVOMDataTransfer" into the Windows start bar search field (Fig. 33). Select the WPI Application (Fig. 34) from the list that appears.



Fig. 33—(Left) Enter EVOMDataTransfer into the Windows search field. Fig. 34—(Right) Select the WPI application from the list that displays.

TIP: After the program has launched, right click on the WPI icon (Fig. 35) in the taskbar and select *Pin to taskbar*. Then, you can double click the taskbar icon to load the program in the future.



Fig. 35—The WPI icon appears in the taskbar when the program is running. Right click to pin it to the taskbar for future use.

2. The EVOM[™] Companion Application opens (Fig. 36).

	-	×
EVOM™ Manual Data Transfer		
Select a folder where you wish to store your data.		

Fig. 36—The application opens and asks you to select a data storage folder.

3. When the EVOM[™] Companion Application is first launched, it prompts you to select a folder for data export. Click on the folder icon in the center of the window. A popup message displays (Fig. 37). Click *OK*.

Pick Export	Folder
Pick the Export F	older to save Plate Files
	Ok

Fig. 37—Select OK to choose a folder to store your EVOM™ Manual data files.

4. A window appears allowing you to navigate to the folder you want to use. Highlight the desired folder and click the *Select Folder* button (Fig. 38).



Fig. 38—Choose the folder you want to use and click Select Folder.

 Once the location is selected, the EVOM[™] Companion Application continuously scans the USB port for an EVOM[™] device and displays a magnifying glass icon (Fig. 39).



Fig. 39—The application is scanning the USB port looking for a connected EVOM™ Manual.

6. Connect your EVOM™ Manual to any USB port on the computer using the USB cable provided (WPI #803026). Plug the USB A type connector into any available port on your computer. See "Appendix F: Installing the EVOM™ Companion Application" on page 49 for installation instructions. Insert the USB type B connector into the port on the back of the EVOM™ Manual. When an EVOM™ Manual is connected, wait for a few seconds for the application to find it. Once found the icon changes to indicate that the EVOM™ Manual was found. It displays the selected directory under the icon (Fig. 40).



Fig. 40—The EVOM™ Manual was found, and data files will be stored in the directory designated at the bottom of the window.

Viewing Data in a Spreadsheet Program

1. If you stored the data on the USB flash drive, remove it from the **EVOM™ Manual** and plug it into a free USB slot in your computer. Using Windows® Explorer, navigate to the USB flash drive directory. If you saved the data directly to the PC, navigate to the designated storage directory.

- 2. In the directory, look for files named *PlateX.csv*. (If you designated a file name or a prefix, your files will be named differently.)
- 3. Double click on a file to open it. If your computer has Microsoft[®] Excel or another spreadsheet program, the file opens. The well data is loaded into spreadsheet cells.
- 4. To transfer the data file to your computer, select the plate files and copy it to a folder on your computer.

Sample data file shows:

- Date, time, plate size
- EVOM™ Manual firmware revision
- Measurement mode with a blank or a voltage offset (PD indicates Potential Difference and is seen when taking voltage measurements.)
- Units (for resistance measurements, ohms or kilo ohms, for voltage measurements mV or V.)
- Recording direction, 1-4 (See "Plate Selection Page and Plate Preview" on page 12.)
- Resistance scale (10K, 50K, 100K), and the last used scale is shown
- Well data in the plate grid with the blank offset applied

Sample Resistance File (*.csv)

Date 03/12/2020 13:11:56 Well Plate 24 EVOM™ Manual Version 3.0 MODE OHMS offset 999.8 Units: OHMS Direction 1 Resistance Scale 50k Ohms

0	1	2	3	4	5	6
A	19015	19015	19015	19015	19015	19015
В	19016	19016	19016	19016	19016	19016
С	19016	19016	19016	19016	19016	19014
D	19014	19014	19014	19014	19014	19017

Sample Voltage File(*.csv)

Date 03/12/2020 13:12:48 Well Plate 24 EVOM™ Manual Version 1.0 MODE PD offset -0.0 mV Units: mV Direction 1 Resistance Scale 50k Ohms

0	1	2	3	4	5	6
A	0.0	0.0	0.0	0.0	0.0	0.0
В	0.0	0.0	0.0	0.0	0.0	0.0
С	0.0	0.0	0.0	0.0	0.0	0.0
D	0.0	0.0	0.0	0.0	0.0	-0.0

USB Testing

From the *Store Options* page you can test a USB flash drive to verify that it can be used. This drive must be formatted as FAT32.

- 1. Insert the drive to see if it is recognized by the **EVOM™ Manual**.
- 2. From the main page, press and hold the *Store* button. Then, press the *USB Test* button.



Fig. 41—Press the Test USB button to run the test.

3. Press the *Test USB* button to run the test or *Return* to abort the test. The USB drive will be written to with 100 lines of data to a file named USBTEST.txt and those lines will be read back. If the drive is usable a message indicates that the USB flash drive can be used (Fig. 42), otherwise an error displays (Fig. 43).

This USB stic	k can be used.	Error OPENING USB file!			
USB Test Cor	nplete!	Error: ERROR -12			
Test USB	Return	Test USB	Return		

Fig. 42—(Left) The USB flash drive passed the test and is suitable for use with the EVOM™ Manual.

Fig. 43—(Right) The USB flash drive is not suitable for use with the EVOM™ Manual.

4. Insert another USB flash drive and press the *Test USB* button to test another drive or press *Return* to go back to the *Store Options* page.

OPERATIONS

IMPORTANT! Before you start making measurements, WPI recommends that you:

- Charge the **EVOM™ Manual** battery or run under mains power.
- Clean the electrodes. Refer to the Maintenance section of the electrode manual for cleaning protocols.
- If it is new, test the USB flash drive. See "USB Testing" on page 24.
- Validate the EVOM™ Manual using 1000 Ω test resistor or calibrate the EVOM™ Manual.

Instrument Diagnostics and Calibration

WPI recommends that **EVOM™ Manual** be put through the diagnostics described below before using it for the first time and then periodically thereafter. If there is a concern that the meter or the electrode is not functioning properly, the following protocols may be used to confirm the **EVOM™ Manual** operating status.

Testing the EVOM™ Manual Meter (Resistance) and Calibration

- 1. Insert the **99673** test resistor into the electrode port on the meter's right side.
- 2. On the main display, select the *Setup* page. In the *Mode units* area, select *Ohms* and 10,000. Press *Return*.
- 3. Verify that the blank handling is disabled (Setup>Blank Handling). The screen should display 1000 $\Omega \pm 1 \Omega$. If it does not, select *Setup*, push the *Calibration* button and wait for the process to complete. (This process takes 30 seconds to complete.) When the date and time are displayed, the calibration process is complete.
- 4. Press *Return* to access the main page. The display should show 1000 $\Omega \pm 1 \Omega$.
- 5. Press the *OK* in the upper left of the octagonal icon on the main display page to short the electrodes. Verify that the **EVOM™ Manual** reads 0 ±1 Ω.
- 6. To return to normal **EVOM™ Manual** operation, unplug the test resistor and press the **■** button (upper left).

Testing the EVOM™ Manual Meter (Voltage)

- 1. On the main display, select the *Setup* page. Press the *Short Electrodes* button.
- 2. In the *Mode units* area, select *Millivolts*. Press *Return*.
- Press the *Probe Null* button and then *Disable*. Press *Return* twice. The main display should show 0.0 mV. If it does not, verify that the probe null is not active and that it has a 0.0 V offset, ±0.1 mV.
- 4. Press the 🕔 button (upper left) to remove the electrode short.

Test Resistor "Calibration Kit"

The **99673** test resistor is supplied to verify the calibration.

Taking Measurements

Configuring Data Storage Options

- 1. Power on the **EVOM™ Manual** using the power switch on the rear panel.
- To configure your data file storage settings, hold the *Store* button on the main screen for 2 seconds or press *Store Screen* on the *Setup* screen to access the Store Options screen.



Fig. 44—Store options screen.

Select the preferred storage mode by selecting one of the radio buttons, *USB Save* or *APP Save*. USB Save stores data files to a USB flash drive, and the APP Save stores the data files on a connected computer using the EVOM[™] Companion Application.

NOTE: Data can only be stored in one location. The same data cannot be stored in both the PC Companion Application and a USB flash drive.

If a filename other than the default (plate1) is required, choose whether you will use a prefix and auto-indexing or a unique file name. To use a prefix and auto-indexing, activate the *Auto Index* radio button. Then, press *Store Prefix* to enter a file name. To use a unique file name, deactivate the *Auto Index* radio button. Then, select *File Name* to enter a new name.



Fig. 45—A keyboard displays so you can enter a unique file name.

- Press Clear.
- Use the on screen keypad to enter a new name.
- Press *Enter*.

NOTE: Auto indexing changes the name to a numeric sequence formatted with the name, a prefix and a sequential number (name_prefixN).

Preparing Storage Location

Prepare the storage location you selected above, either USB or PC using the appropriate method:

- If you selected USB Save, you need to insert the USB flash drive into the USB port on the side of the EVOM[™] Manual. See "Storing Data on the USB Flash Drive" on page 17.
- If you selected *APP Save*, you need to make sure the EVOM[™] Manual is properly connected to the computer using the USB cable included and verify that the Companion Application is running. See "Storing Data on a Connected PC" on page 19.

Configuring Plate Settings

Before taking measurements, you need to establish the settings for your plate, including the number of wells, the mode (resistance or voltage) and the mode units, and prepare the electrodes. Select either the Resistance Configuration Procedure or the Voltage Configuration Procedure below.

Resistance Configuration Procedure

If you want to take resistance measurements, configure your plate as follows:

1. Press the *Setup* button on the main screen to configure your plate settings.



Fig. 46—Press Setup to access the Setup screen.

- 2. Press the *Select Plate* button to choose the plate size. See "Plate Selection Page and Plate Preview" on page 12.
- 3. Press the *Mode Units* button and choose either OHMS or KOHMS for making resistance measurements (Fig. 47).
- 4. Select the resistance range (Auto, $100K\Omega$, $50K\Omega$ or $10K\Omega$) and press the *Return* button (Fig. 47).



Fig. 47—*Select the mode. For resistance measurements, choose OHMS or K OHMS. Then select the range.*

- 5. Press the *Calibrate* button to calibrate the electrode. (This uses a 10 K Ω 0.1% resistor). See "Instrument Diagnostics and Calibration" on page 25.
- 6. To subtract a blank well (including the fluid and electrode resistances) press the *Blank Handling* button. Then, place the electrode in a blank well and press the *Reading* button. Press the *Blank Enable* radio button to subtract the blank value. Press the *Return* button (Fig. 48).



Fig. 48—(Left) Blank Handling window.

Fig. 49—(Right) The main screen shows the offset underneath the reading.

7. Press *Return* to navigate back to the main screen (Fig. 49).

Voltage Configuration Procedure

If you want to take voltage measurements, configure your plate as follows:

1. Press the *Setup* button to configure your plate settings.



- *Fig.* 50—(*Left*) *Press Setup to access the Setup screen.*
- 2. Press the *Select Plate* button to choose the plate size. See "Plate Selection Page and Plate Preview" on page 12.

 Press the *Mode Units* button and choose either Millivolts or Volts for making voltage measurements (Fig. 51). Press the *Return* button to go back to plate settings screen (Fig. 50).



Fig. 51—(Right) Select the mode. For voltage measurements, choose Millivolts or Volts.

4. To calibrate (zero) the electrodes' galvanic charge in saline, place the electrodes in a saline solution, and press the *Probe Null* button. A popup message displays (Fig. 52). Push the *NULL* button. Wait for the second message stating "Solution value 0.0" before pressing the *Return* button. This action removes any voltage offsets on the electrodes. Then, activate the *Enable/Disable* radio button to offset the electrode potential.



Fig. 52—(Left) Null probe process.

- Fig. 53—(Right) The potential difference offset shows on the main screen under the reading.
- 5. Press *Return* to navigate back to the main screen (Fig. 53).

Taking Measurements

Whether you are taking resistance or voltage measurements, the data acquisition procedure is identical.

- 1. To begin measuring with well A1, do one of the following:
 - Press the foot switch or touch the *Store* button on the main screen. A
 measurement of the first well is taken, and the data storage advances to the
 next well in your sequence. Tap the *Store* button again to measure the next
 well in your defined sequence.

NOTE: When required, you may use the *Previous Well* area on the left side of the screen to return to the last well and re-measure or the *Next Well* area on the right side of the screen to skip a well. Then, press the *Store* button or the foot switch to take the measurement of that cell.

Press the *Preview* button to see a graphical display of the plate. The green cell
indicates the cell to be sampled, and the yellow cell indicates the next well in

the sequence. Press the foot switch to take a measurement and advance to the next well (Fig. 54).

2. Repeat the sampling process. Press the *Preview* button to see the measurements that have been taken (Fig. 54).



- *Fig.* 54—The preview screen for a 6-well plate show the A1-A3 cell data taken (1000).
- 3. Once the last well has been recorded, the file storing selection notification appears prompting you to take an action (Fig. 55 and Fig. 56).



Fig. 55—(*Left*) *After the last cell measurement is taken and you are saving to the USB drive, this message appears.*

Fig. 56—(Right) If you are saving to the EVOM™ Companion Application, the message changes a little.

Select one of the options as follows:

- *Export* starts the storage of all the plate files. When you select it a message appears describing which plate is being stored (Fig. 57). The number of plates could be one, two or three, depending on how many were collected before exporting.
- Next is available on plate 1 and 2, but not on plate 3. It returns you to the main screen so you can take measurements of the next plate which starts from a clear state. The note above the Setup button on the main screen indicates which plate data is being collected (Fig. 5).

NOTE: If you are on plate 3, the *Next* button does not appear, because EVOM™ Manual's memory only holds data from three plates.

• **Back** returns you to the main screen (Data Collection). You go back to the first well of the current plate being collected and can acquire data again.



Fig. 57—(Left) The message changes showing which plate is being saved. Here Plate 3 data is being stored on the connected computer.

Fig. 58—(*Right*) All three plates were saved to the connected computer.

4. When the plate data has been saved, a confirmation message appears (Fig. 58). Press the *Done* button to return to the main screen.

NOTE: If you save to the USB flash drive and the drive is not plugged in or it is not recognized, a warning message appears (Fig. 59). Press the *Back* button and try another USB flash drive. The Export function cannot be used on an unrecognized drive.



Fig. 59—When the USB flash drive is not plugged in or not recognized, a warning message appears.

NOTE: **EVOM™ Manual** should respond with "Plate data saved to USB" within 15 seconds. If "Saving file..." is still seen after 20 seconds, press the *Back* button and use another USB flash drive. Once a file has been saved, open the file on a computer to verify the contents.

- 5. Now you can work with your data.
 - If you stored your data on the USB flash drive, remove the drive and transfer the data files to your computer. They are in comma separated value (CSV) file format for Microsoft[®] Excel.
 - If you stored your data using the EVOM[™] Companion Application, all your data files will be located in the directory you specified when you started the Companion Application.

SYSTEM MESSAGES

Here are some warning messages you may see.

Last Cell Warning

When you are taking measurements, and you reach the last cell, a message appears.



Fig. 60—(Left) After the last cell measurement is taken and you are saving to the EVOM™ Companion Application, this message appears.

Fig. 61—(*Right*) *If you are saving to the USB drive, the message changes a little.*

These messages indicate that the last well in the plate has been reached, and an action on your part is required to proceed.

- *Export* starts the storage of all the plate files. When you select it a message appears describing which plate is being stored (Fig. 60). The number of plates could be one, two or three, depending on how many were collected before exporting.
- Next is available on plate 1 and 2, but not on plate 3. It returns you to the main screen so you can take measurements of the next plate which starts from a clear state. The note above the Setup button on the main screen indicates which plate data is being collected (Fig. 5).
- *Back* returns you to the main screen (data collection). You go back to the first well of the current plate being collected and can acquire data.

Power Level Messages

When the battery level falls below 5%, a warning message appears.



Fig. 62—The battery level is below 3%.

When the battery power level falls to 1%, a warning message appears. Save your data file. The data is saved to a backup file named PWRFAILDATA.TXT.

Low battery. Saving backup data. Please wait... Low battery. Connect Power! Plates SAVED 3

Fig. 63—(*Left*) When the battery level falls to 1%, you are asked to save your data file. *Fig.* 64—(*Right*) Once the data is save, this message appears.

When the battery level falls to 0%, the system begins a 60 second countdown before powering off.



Fig. 65—When the battery power falls to 0%, a warning displays briefly. *Fig.* 66—The 60 second countdown was initiated when the battery power fell to 0%.

Before shutdown, a final warning appears.



Fig. 67—Immediately before the system powers off, the shutdown message displays.

Reloading a Backup File after Power Failure

NOTE: This feature is only available in the USB save mode. When using the EVOM[™] Companion Application, individual .CSV files are created for each plate in memory and are in the same folder where all other .CSV files are saved using the file naming convention designated on the Store Options screen.

1. When power is restored a green message appears (Fig. 68). Press OK.



Fig. 68—(Left) Power level is restored when the unit is plugged into a power source.

2. Cycle power on the **EVOM™ Manual** by turning it off and on again by sliding the power switch on the back panel. This automatically loads the backup files which

were saved when the power failed. The backup files are deleted from the USB flash drive. You will see messages that indicate which plate or plates are being restored (Fig. 69). Then, a success message appears (Fig. 70). Press the *OK* button to preview the data.



Fig. 69—(*Left*) *The backup file is restored on power up.*

Fig. 70—(Right) When the files are restored, this message appears. Press OK to preview the data.

3. Press the *Store* button on the main screen to save the data.

ACCESSORIES

WPI Part #	Description
EVM-EL-03-01-01	EVOM™ EndOhm-6 Electrode for TEER in 6 mm Insert
EVM-EL-03-01-02	EVOM™ EndOhm-12 Electrode for TEER in 12 mm Insert
EVM-EL-03-01-03	EVOM™ EndOhm-24 Electrode for TEER in 24 mm Insert
EVM-EL-03-02-01	EVOM™ STX HTS Electrode for TEER in Corning 96
EVM-EL-03-02-02	EVOM™ STX HTS Electrode for TEER in Milipore 96
EVM-EL-03-02-03	EVOM™ STX HTS Electrode for TEER in Corning Biocoat/Falcon 24
EVM-EL-03-02-04	EVOM™ STX HTS Electrode for TEER in Corning Costar 24
EVM-EL-03-03-01	EVOM™ STX4 Electrode with Removable Blades for TEER in 6 mm Inserts
EVM-AC-03-01-01 EVM-AC-03-03	EVOM™ Electrode Blades for the EVOM™ STX4 for TEER, 3 pairs EVOM™ Warming Plate

TROUBLESHOOTING

EVOM™ Manual Meter: Most of the time, system problems are related to the electrode, not the meter itself.

Some issues you may encounter include:

- UNSTABLE OR HIGH READING: If the electrode fails, the most common symptom is an unstable or unusually high reading.
- LOW RESISTANCE: When the meter displays a lower than expected resistance, but is stable and reproducible, the most likely cause is related to the cell culture, not necessarily the meter or the electrodes.

Issue	Possible Cause	Solution		
ind off	Unit is	Recharge the unit for at least 12 hours.		
Unit rur briefly, a powers	insufficiently charged	NOTE: Make sure the EVOM™ Manual is powered off when charging the battery.		
oes not r charging not lit	Insecure connection at power input jack on meter	Verify that the power connector is securely connected at the meter power input jack.		
Unit d charge o LED	Defective charger	The EVOM™ Manual charge socket can be tested with a USB to Mini-B cable plugged into a computer's USB port (WPI #803026). If the charging LED lights up, then a new charger is needed (WPI #803025).		
USB drive not working /not recognized	Not all USB flash drives work on the EVOM™ Manual .	See "Sample Resistance File (*.csv)" on page 23 and page 5.		
er will not power n with charger SCONNECTED	Batteries are discharged	The meter powers down automatically when the batteries are depleted. Power off the meter and connect it to the battery charger. Charge it for at least 12 hours. A full charge may require up to 24 hours. For reliability, the meter should be charged each evening (with the power off) before use the next day.		
DO	Batteries are defective	Contact WPI for service. The EVOM™ Manual battery is not user serviceable.		

Issue	Possible Cause	Solution			
eading is unstable	Electrodes are heavily charged or dirty	Clean the electrodes as per the instruction manual for the electrode.			
	Electrode too close to strong electromagnetic radiation device	Move the system to a different area away from sources of electromagnetic fields. Electromagnetic field sources could include computers, MRI equipment, magnetic stirrers, etc. Cell phone signals can also interfere.			
Voltage r	Power line or USB connected to a recording device causing noise	Remove the USB connection to eliminate the possibility of a ground loop.			
e set near zero	Electrode not equilibrated	The Ag/AgCl electrodes (inner voltage measurement electrodes) on the probe may become polarized due to an uneven distribution of ions between the electrode pair. This ionic charge can be neutralized by immersing the probe into 0.1M KCL for 24 hours with the probe electrodes shorted electrically. The probe is shorted electrically when it is connected to the meter with the power OFF, and the FUNCTION switch in the OHMS position. See "Probe Null" on page 15 for information on how to null the electrodes.			
n not k	Electrode requires cleaning	Clean the electrode as per the instructions manual for the electrode.			
Voltage reading is can	Contamination between electrodes	Inspect the inter-electrode surface areas for material which could form an insulative coating on the surface of the inner and outer electrodes. If the material cannot be removed, the electrode should be replaced.			
	Corrosion has formed on conductive traces	An insulative, transparent coating protects the copper conductors that connect each electrode to the main wiring in the handle. When the probe is very old, this coating can deteriorate, allowing the salts in the media solution to leech through to the copper conductors. This is observed as a black discoloration on the copper circuit traces underneath the conformal coating. If this is observed, replace the electrode.			
Resistance reading unusually high	Contaminated electrode contacts	With use, the chopstick electrode probe contacts may acquire a buildup of protein or other foreign material that effectively increases the baseline resistance of the electrodes. The electrodes can be cleaned using various methods.			

Issue	Possible Cause	Solution
Resistance reading unusually low	Cell culture or media problem	If the cell culture has been given sufficient time to achieve confluence, and the reading is stable but significantly lower than expected, then the problem is probably related to the cell culture. Electrode failure will not generally cause a lower than expected yet stable reading. Use the test resistor to verify the meter is functioning correctly. The meter display should read 1000Ω . Refer to the appropriate electrode manual for instructions on testing the electrode. See "Appendix C: Improving the Accuracy and Repeatability of the System" on page 45.
Instable	Electrodes are not fully immersed in culture media solution	The meter will not provide a stable reading if the electrodes are disconnected from the meter or if the electrodes are not fully immersed into culture media.
Resistance reading drifts or is u	Electrodes are not held still during measurement	Handheld electrodes must be kept as motionless as possible during a measurement. Excessive movement will cause the measurement to fluctuate.
	Charger is connected to the meter	The meter reading can become unstable due to the loss of electrical isolation when the charger is connected to the AC power. To ensure stability of readings, always disconnect the charger from the meter when making measurements.
	Old electrode	Use the test resistor to verify the meter is functioning correctly. The meter display should read 1000Ω . The lifetime of the electrodes is between 1- 2 years with normal use. See the previous note on corrosion and the note on coatings.
nce is a negative value	Contamination on electrodes	Use the 1000Ω test resistor to verify that the meter is working correctly. The meter display should show 1000Ω . If a negative value exists only when using the STX electrode, inspect and test the electrode for the possible formation of an insulative coating of foreign material that electrically blocks the current and voltage electrodes. Remove the foreign material and retest. Refer to the appropriate electrode manual for instructions on testing the electrode.
Resistal		It a red negative value shows on the display only when using the electrode and the blank setting is off or at zero, then inspect the electrode for the formation of an insulative coating of foreign material that electrically blocks the current and voltage electrodes.

Issue	Possible Cause	Solution				
The reading appears to be drifting* (see the drift definition below).	The electrodes are coated with deposits from the use of media or buffer.	The electrodes require enzymatic cleaning.				
	In a 5% CO_2 environment, a loss of CO_2 causes the media pH to change, and the resistance reading may increase sharply.	A 5% CO $_2$ environment can help in reducing pH media changes.				
The electrode reading is unstable. (See the stability definition below).	The electrodes are coated.	The electrodes require enzymatic cleaning.				
	Radio frequency interference	Turn off or move any cellular phones farther away from the experimental setup.				
appears on the screen	Resistance reading is out of range	An out of resistance range will display on the screen. Adjust the range to correct the error. A saved well will read a space ("") if the display is out of range. Go to <i>Setup, Mode Units</i> and increase the resistance range. See page 11. NOTE : The EVOM™ Manual will read only about 5% above				
1		100Κ Ω.				

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 wpi-ts@wpiinc.com.

*Drift-Readings that continuously increase or decreases a significant value (either voltage or resistance) over time. Example: At 1000 Ω the reading is increasing 100 Ω /minute. (A drift of 10 Ω /minute is acceptable.) Excessive drift may be caused by unwanted deposits on the electrode, pH changes or temperature variance.

Instability–At 500 Ω , the reading jumps from 450 to 550 Ω and will not settle down (an instability ±5 Ω is acceptable in the 500 Ω range. In the higher ranges, up to ±1000 Ω is acceptable at the 100K range. Electrodes showing instability may be coated with media or buffer constituents and may require enzymatic cleaning.

SPECIFICATIONS

This unit conforms to the following specifications: Tissue Sampling Frequency12.5 Hz 0 to 50,000 Ω0 to 100,000 Ω +5% Auto Mode1 to 100,000 Ω auto current 2 μ A, 4 μ A, 10 μ A Voltage Resolution......0.001 V, 0.1 mV Accuracy Voltage......± 0.1 mV Auto mode 1 to 100,000 Ω auto current 2 μ A, 4 Display Update Rate0.5 seconds Certifications......CE Firmware......Upgradeable* *NOTE: A USB to Mini-B cable (WPI #803026) is required along with PC bootloader software and the image are required to upgrade the firmware. **Not user serviceable. Contact WPI for repair or replacement.

APPENDIX A: FILE FORMATS

Resistance (*.CSV)

Date 03/12/2020 13:11:56 Well Plate 24 EVOM[™] Manual Version 2.0 MODE OHMS offset 999.8 Units: OHMS Direction 1 Resistance Scale 50k Ohms

0	1	2	3	4	5	6
A	19015	19015	19015	19015	19015	19015
В	19016	19016	19016	19016	19016	19016
С	19016	19016	19016	19016	19016	19014
D	19014	19014	19014	19014	19014	19017

Voltage (*.CSV) Date 03/12/2020 13:12:48 Well Plate 24 EVOM[™] Manual Version 2.0 MODE PD offset -0.0 mV Units: mV Direction 1 Resistance Scale 50k Ohms

0	1	2	3	4	5	6
A	0.0	0.0	0.0	0.0	0.0	0.0
В	0.0	0.0	0.0	0.0	0.0	0.0
С	0.0	0.0	0.0	0.0	0.0	0.0
D	0.0	0.0	0.0	0.0	0.0	-0.0

APPENDIX B: CONTINUOUS RECORDING VIA USB (PYTHON) ON A PC

The program on the USB flash drive is a self-executing program to record serial data through a USB cable to a Window PC computer. This is intended to record continuous measurements of a single sample. The following files are included on the USB stick:

- CDM v2.12.28 WHQL Certified EVOM™ Manual USB driver for Windows
- Python.exe Python recording program and other support folders for Windows
- Read_EVOM_HOMECNX.bat Batch file for executing the recording program
- WPI_EVOMCNX_5C.py Python EVOM™ Manual USB digital recording program
- WPI_EVOM_Manual.csv Sample recording file

NOTE: An out of resistance range indication on the **EVOM™ Manual** screen will show -----. The Python output will list -99999.9. If you are using a lower range (10K or 50K), stop the output (X), increase the range and restart. The maximum resistance level that the **EVOM™ Manual** can measure is 10,5000 ohms.

Running the External Data Connection Output Program (PC, Mac, Linux)

Windows 8 or above users:

WPI provides a USB keydrive (also known as a flash drive) with all the software needed to run the External Data Output program. To use these programs:

1. Connect the **EVOM™ Manual** to the USB port of the PC. The active remote connection port for the **EVOM™ Manual** service port is in the back of the unit as a USB-B connector [marked in red].



Fig. 71—Plug the USB-B (red) connector into the port on the back of the EVOM[™] Manual. The plug-and-play service from Microsoft should install the driver automatically, however if a driver is not found, there is a driver in the folder on the USB drive called CDM v2.12.28 WHQL Certified, which can be used install the driver.

- 2. Plug the USB flash drive into a USB slot on your PC. If your PC does not have a spare USB slot, you will have use the Windows option [B] below.
- 3. Open a Windows Explorer window and navigate to the USB flash drive folder on

the PC. Run the bat file "WPI_EVOMCNX_3.E.bat." The program will query the COM ports in use and inform you as to which port the **EVOM™ Manual** is connected.

- 4. Enter the filename for the Output File (ENTER for WPI_**EVOM™ Manual**.csv). Type in a filename and add the .csv extension or enter to accept the default.
- 5. Enter the time between readings in seconds. (Press *Enter* for minimum delay.) Enter a time of 0, 1, 2 or 5 seconds. (0 is fastest at about 0.3 seconds.) The program will run and update until ^X (control X) is typed. The readings are continuous at 12.5 Hz, always on. The time entered is the averaged reading taken at that time interval.
- 6. The default file name is WPI_**EVOM™ Manual**.csv. This file may be opened in Microsoft[®] Excel.

For Windows users that can not use a USB keydrive, or only have one USB port, or for Linux OS or Mac OS users

- 1. For Windows, Linux or Mac users that already have Python 3.8.0 or higher installed, skip to step 2. If you do not have Python installed, download and install Python for your operating system from www.python.org . Be sure to download the version appropriate for your operating system. The minimum supported Python version is Python 3.8.0
- 2. Make sure all the Python libraries are available as keyboard, serial and glob. From a command line window, run these commands (all lower case) :

pip install glob pip install serial pip install keyboard

- 3. Download the file **EVOM™ Manual** digital output archive for your operating system from https://www.wpiinc.com/support/software-download/. This archive has all the Python program files to use the external data output.
- 4. Extract the files to a WPI directory or a directory of your choice.
- 5. Connect the **EVOM™ Manual** to the USB port of the PC. The active remote connection port for the **EVOM™ Manual** service port is in the back of the unit as a USB-B connector [marked in red].



Fig. 72—Plug the USB-B (red) connector into the port on the back of the EVOM™ Manual.

- 6. Open Python or a command window on your system.
- 7. At a command line or from python type this command : python WPI_ EVOMCNX_3.E.py and press enter. If you are not running Python from the directory where WPI_EVOMCNX_3.E.py was extracted, you will have to change directory to that location.
- 8. Enter the filename for the Output File (ENTER for WPI_EVOM_Manual.csv). Type in a filename and add the .csv extension or enter to accept the default.
- 9. Enter the time between readings in seconds. (ENTER for minimum delay.) Enter a time of 0, 1, 2 or 5 seconds. (0 is fastest at about 0.3 seconds.) The program will run and update until ^X (control X) is typed. The readings are continuous at 12.5 Hz, always on. The time entered is the averaged 20s/s reading taken at that time interval. Note: 2 seconds is recommended.
- 10. The default file name is WPI_EVOM_Manual.csv. This file may be opened in Microsoft® Excel or any other spreadsheet program as a .CSV file



Sample CSV Recording

TIME , DATA , VALUE 12 Mar 2020 13:24:35,0,0000.0 12 Mar 2020 13:24:36,1,19017.9 12 Mar 2020 13:24:36,2,19018.1 12 Mar 2020 13:24:36,3,19018.0 12 Mar 2020 13:24:37,4,19018.1 12 Mar 2020 13:24:37,5,19017.8 12 Mar 2020 13:24:37,6,19017.7 12 Mar 2020 13:24:37,7,19017.7 12 Mar 2020 13:24:38,8,19017.8

NOTE: Python is an open source programming language. Licensing information can be found on the Python website (https://docs.python.org/3/license.html#terms-and-conditions-for-accessing-or-otherwise-using-python.)

APPENDIX C: IMPROVING THE ACCURACY AND REPEATABILITY OF THE SYSTEM

Whenever possible, operate the **EVOM™ Manual** meter with the battery charger disconnected. This ensures maximum electrical isolation for the most accurate readings. When the charger remains connected to the meter during measurements, electrical noise can be introduced into the system. For the highest accuracy in measurement, WPI recommends disconnecting the unit from the AC power source.

Choose an electrode based on your application (size of the inserts: 6 mm, 12 mm, 24 mm and transwell type).

- **EndOhm** electrodes by design are considered more accurate in capturing smaller changes. However, cell culture inserts need to be physically moved out of the plate to the **EndOhm** chamber for measurement.
- **STX HTS** electrodes are ideal for high throughput screening (HTS) plates.
- With STX electrodes, you can measure inside the well plate. The STX4 electrode is our new and improved version of the STX2-PLUS electrode. It has removable blades.

Fig. 73 demonstrates how the EVOMTM electrodes measures TEER. With a fixed centered electrode geometry of the **EndOhm**, the variation of readings on the same sample is 1-2 Ω , as compared to 5% of the total reading using the **STX2** electrodes. An **EndOhm**, together with the **EVOMTM Manual**, offers the most accurate, convenient and economical solution for trans membrane electrical resistance measurement. For automation of TEER measurements, consider the **REMS** AutoSampler that integrates a robotic system and computer interface to automatically measure tissue resistance in 24- and 96-well plates.



Fig. 73—This comparison shows how the electrode geometry can affect readings.

APPENDIX D: RESISTANCE CALCULATIONS

Resistance

The value of the blank (control well) always adds to the total resistance measured across a tissue culture membrane. The blank resistance must be measured and then subtracted from the resistance reading across the tissue in order to obtain the true tissue resistance. For example, suppose the resistance through a 0.15 M KCl solution and across the membrane support (with no tissue present) of a 12-well cell culture insert measures 130 Ω . This is the blank or control reading for that cell culture insert. (Resistance may vary for culture cups made by other manufacturers.) In this example, using 800 Ω as the sample measurement, the calculated resistance for the tissue itself (R tissue) is:

$$\begin{split} & \mathsf{R}_{\mathsf{Total}} = 800 \ \Omega \\ & \mathsf{R}_{\mathsf{blank}} = 130 \ \Omega \\ & \mathsf{R}_{\mathsf{blank}} + \mathsf{R}_{\mathsf{true \, tissue}} = \mathsf{R}_{\mathsf{Total}} \\ & \mathsf{R}_{\mathsf{true \, tissue}} = \mathsf{R}_{\mathsf{Total}} - \mathsf{R}_{\mathsf{blank}} \\ & \mathsf{R}_{\mathsf{true \, tissue}} = 800 \ \Omega - 130 \ \Omega = 670 \ \Omega \end{split}$$

Unit Area Resistance

As the resistance is inversely proportional to the area of the tissue, instead of reporting resistance, typically the product of the resistance and the area is calculated and reported. The unit area resistance is independent of the area of the membrane used and may be used to compare data obtained from inserts of different sizes.

The unit area resistance is obtained by multiplying the meter readings by the effective surface area of the filter membrane. The dimension is Ω -cm². The resistance is inversely proportional to the surface area. Thus, the larger the membrane, the lower the resistance.

Resistance of a unit area = Resistance (Ω) x Effective Membrane Area* (cm²)

* See manufacturing specifications for the particular insert

Unit Area = 1 cm²

The unit area resistance is independent of the area of the membrane used and may be used to compare data obtained from inserts of different sizes.

Continuing with the previous example, in which the $R_{true tissue} = 670 \Omega$, if an effective membrane diameter (d) was 6.5 mm, the unit area resistance would be:

Resistance x Effective Membrane Area = 670 Ω * π r²

= 670 Ω * 3.14* 3.25* 3.25 / 100

= 670 Ω * 0.331 cm²

= 221.77 Ω-cm².

221.77 Ω is the resistance of a unit area of 1 cm² (TEER).

The larger the membrane, the lower the resistance. The dimension is Ω -cm², not Ω / cm².

TEER Calculations

TEER is a normalized value of resistance per 1 square centimeter of unit area. To compute TEER, multiply the measured resistance by the surface area's listed below.

For example, a 6.6 mm insert measures 1707 Ω , the TEER is 1707 Ω * 0.331 or 565 Ω .

- 6 well plate (24 mm inserts) 4.53 cm²
- 12 well plate (12 mm inserts) 1.13 cm²
- 24 well plate (6.5 mm inserts) 0.3316 cm²
- 96 well plate (4.3 mm inserts) 0.145 cm²

APPENDIX E: FREQUENTLY ASKED QUESTIONS

1. When do researchers want to use the blank function?

Answer: The blank function allows researchers to subtract out resistance measurement that is not caused by the cell layer. This could be a reading affected by the electrodes or fluid resistances or other factors. Normally, you would measure a blank insert with the same volumes as the insert and keep subtracting that blank value from the samples. This way, any offset contributed by the electrode is nullified. The subtracted value is the resistance contributed by the cellular layer.

3. Does the system automatically calculate TEER?

Answer: No. Calculating TEER requires an area calculation. See "TEER Calculations" on page 2 and "Appendix D: Resistance Calculations" on page 46.)

4. The program is automatically stored when the last well is reached. What happens when I only want to measure 8 of 96 wells and store the program?

Answer: Once you're done measuring, open the settings, press the store screen menu, then store the plate data to the USB.

APPENDIX F: INSTALLING THE EVOM™ COMPANION APPLICATION

1. Locate the USB drive (WPI **#97893**), which contain the EVOM[™] Companion Application program and the Python 3.8 program for continuous digital monitoring.

NOTE: The installation package is also available from the WPI website (<u>https://www.wpiinc.com/support/software-download</u>).

- 2. Plug the USB drive into the PC where you want to install the EVOM™ Companion Application.
- Using Windows[®] Explorer, navigate to [DRIVE]:/EVOM_APP_INSTALLER/ directory on the USB drive
- 4. Double click on the *setup.exe* file to install the application. Follow the on screen installation prompts.
- 5. Launch the EVOM[™] Companion Application by typing "EVOMDataTransfer" into the Windows start bar search field (Fig. 74). Select the WPI Application (Fig. 75) from the list that appears.



Fig. 74—(Left) Enter EVOMDataTransfer into the Windows search field. *Fig.* 75—(Right) Select the WPI application from the list that display.

TIP: After the program has launched, right click on the WPI icon (Fig. 76) in the taskbar and select *Pin to taskbar*. Then, you can double click the taskbar icon to load the program in the future.



Fig. 76—The WPI icon appears in the taskbar when the program is running. Right click to pin it to the taskbar for future use.

DECLARATION OF CONFORMITY



WORLD PRECISION INSTRUMENTS, LLC. Telephone: (941) 371-1003 Fax: (941) 377-5428 e-mail <u>wpi@wpiinc.com</u>

DECLARATION OF CONFORMITY CE

We:

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

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

EVM-MT-03-02

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

Low Voltage Directive (Safety) 2014/35/EU:

• EN 61010-1:2010+A1:2019

EMC Directive 2014/30/EU:

- EN IEC 61326-1:2021
- EN IEC 61326-2-3:2021
- EN IEC 61000-3-2:2019+A1:2021
- EN IEC 61000-3-3:2013+A2:2021

Robert Lewis / Director of Design and

Development

Issued On: January 21, 2025

Europe Representative Mr Andrew Waldes Managing Director World Precision Instruments Germany GmbH, Pfingstweide 16, 61169 Friedberg, Germany

F-QC-006 Rev D

WARRANTY

WPI (World Precision Instruments) 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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