

User Guide

Millicell® ERS 3.0 Digital Voltohmmeter

Electrical Resistance System

MERS03000



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Introduction

The Millicell® ERS (Electrical Resistance System) 3.0 Digital Voltohmmeter is an instrument and electrode probe designed to reliably measure Trans-Epithelial Electrical Resistance (TEER) of epithelial cells in culture. A change in TEER detected with the electronic circuit of the Millicell® ERS 3.0 Digital Voltohmmeter and its electrode probe is an indication of cell monolayer health and confluence.

Hard gold plate on the interior and exterior facing surfaces of the electrode measures voltage. The small size of the electrode enables the user to easily measure trans-epithelial voltage and the resistance of cells grown on microporous membranes.

This system is for research use only.

Millicell® ERS 3.0 Digital Voltohmmeter and Components

The Millicell® ERS 3.0 Digital Voltohmmeter consists of an instrument, standard electrode probe, verification adapter, wireless network dongle, and power/charge cable.

Instrument Base

The interface for the instrument is an intuitive touch screen. The back panel of the instrument has three USB ports, an Ethernet port, a foot-switch port, electrode probe port, toggle off/on switch and a power cable port.



Benefits Using AC Power Source

The Millicell® ERS 3.0 Digital Voltohmmeter uses alternating current (AC) to make membrane resistance measurements. Using an isolated AC source has several advantages over traditional use of direct current (DC):

- Membrane voltage and voltage electrode offset do not affect resistance measurements.
- There is no electrochemical deposition of electrode metals.
- Membrane capacitance does not affect resistance readings. Once the Millicell® ERS 3.0 Digital Voltohmmeter is standardized, you can use it to quantitatively measure cell confluence.



Performance Specifications

Membrane Voltage Range	± 200.0 mV
Voltage Resolution	0.1 mV
Voltage Accuracy	± 2 mV
Resistance Range	0-100,000 Ω
Resistance Resolution	1 Ω
Resistance Accuracy	≤ 1% accuracy
Resistance (auto ranging)	10 μA (0-10K Ω) nominal at 12.5 Hz
AC square-wave current	4 μA (10K-40K Ω) nominal at 12.5 Hz
	2 μA (40K-100K Ω) nominal at 12.5 Hz
Thermistor Dynamic Range	68-98.6 °F (20-37 °C)
Temperature Accuracy	+/- 1 within 68-98.6 °F (20-37 °C)
	59-86 °F (15-30 °C)
Environmental range	20-80% non-condensing relative humidity
	For indoor use only
Dimensions	H: 3.35 inches (8.5 cm) W: 8.8 inches (22.4 cm) D: 5.55 inches (14 cm)
Weight	1.77 lbs. (0.805 kg)
Instrument Body Housing Material	ABS Plastic
Display Resolution	1024 x 600 pixels

Standard Electrode Probe

The electrode probe is connected to a shielded cable with a MiniDIN plug at its end. This plug connects to the Sensor port at the back of the instrument during use.



Instrument Power Supply/Charge Cable

The 60 W single output power supply can be used directly with the instrument with or without the battery present.

Specifications for input and output ratings are provided in the Safety Sheet shipped with the product or from the product page at SigmaAldrich.com.



USB Wireless Network Dongle

The external USB Wireless network dongle provides wireless connectivity on 5GHz band reducing wireless interference. The dongle will send data from the instrument to the Millicell® Cloud.

The USB Wireless network dongle can be plugged into any of the instrument's three USB ports. It can stay plugged in during storage, usage, and charging.



Millicell® ERS 3.0 Verification Device

The verification adapter is an essential part of the Millicell® ERS 3.0 because it informs the user that their device is measuring Resistance and Temperature within appropriate ranges. A failure to read the verification adapter values within acceptable ranges is indicative of something wrong with the instrument or software.



Battery Pack

(sold separately)

Millicell® ERS 3.0 Digital Voltohmmeter uses a rechargeable, smart, lithium-ion battery pack. (14.4 V/3.45 Ah/49.70 Wh)



When the battery power level falls below a minimum threshold, the instrument automatically powers off. A fully charged battery will provide power for approximately 4 hours of continuous usage.

Battery Power	Internal 14.4 V Li-Ion 3450 mAh 49.68 Wh
Nominal battery run time	Approximately 4 hours
Weight (instrument with battery installed)	2.3 lbs. (1.035 kg)

Note: The instrument can be utilized while the battery is charging. However, voltage measurements are more stable on battery power only (unplugged from wall power).

Foot Pedal

(sold separately)

Ideal for hands free operation of the instrument to save an on screen value as the user moves the electrode probe to the next measurement well.



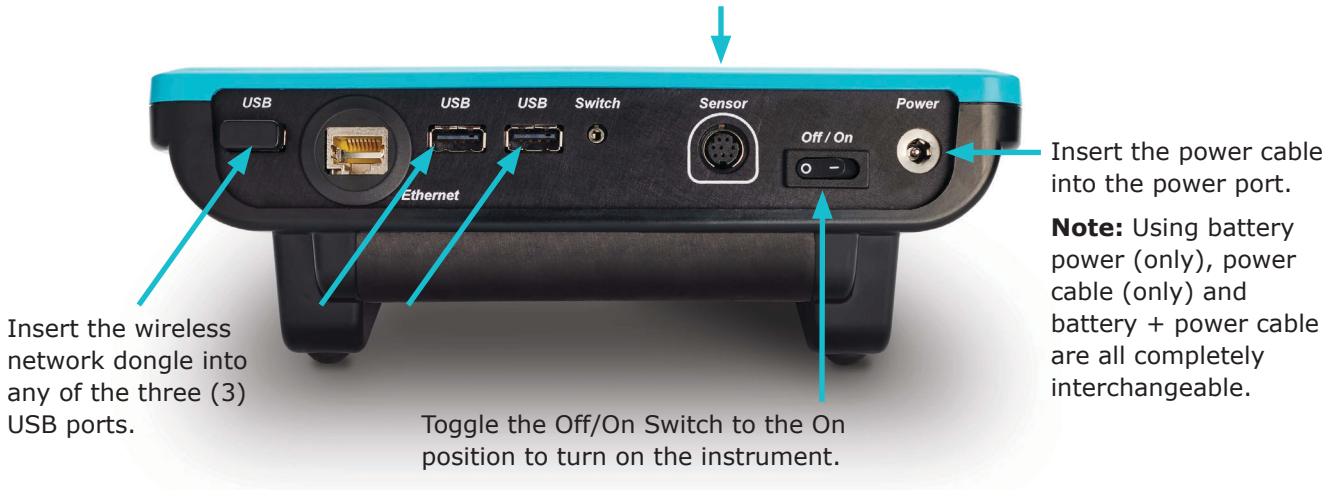
Set Up

Instrument

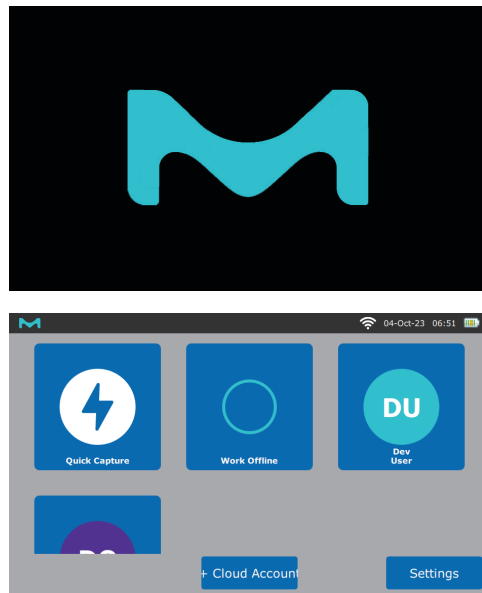
From the back of the instrument:

Insert the Standard Electrode Probe into the Sensor port.

















Note: The Mini-DIN plug pins should line up with the port holes to ensure that neither is damaged. There is an outline of the plug drawn into the back panel for easier alignment. The flat side of the connector should be facing downward.



The M logo will appear while the instrument is initializing. When ready, the main menu screen will be displayed.



Software Symbols

Symbol/ Button	Definition	Symbol/ Button	Definition
	Back to previous screen		Calendar
	Home/Log out button which returns user to the main menu. The main menu will have each Millicell® Cloud account user, offline, and quick capture options.		Information
	Wireless network connected		Locked
	Ethernet connected		Failed operation
	From a selected plate, this signifies the option to adjust the plate name and seeding date.		Battery status (shows full)
	Uploading data		Battery half full
	Upload data error, upload cannot be completed		Battery low
	Checkmark		Battery depleted, charge required

Insert Plates

Recommended media volumes for standard plastic culture plates with Millicell® hanging inserts:

Insert	24-well	12-well	6-well
Well Diameter (mm)	6.5	12	24
Membrane surface area (cm ²)	0.3	1.1	4.5
	100	200	1000
Apical volume (µL) for Millicell® inserts	200	400	2000
	300	600	3000
	400	800	4000
	600	900	2000
Basolateral volume (µL) for cell culture plates	900	1200	2750
	1200	1500	3500
	1500	1800	4300

← Recommended Volume

← Recommended Volume

Note: Plates vary slightly. Volumes should be adjusted as required.

Recommended Greiner Bio-One CELLSTAR Cell Culture Multiwell 6-, 12-, and 24-well plates for use with the 24-well Millicell® hanging inserts.

Apical and Basolateral Medium Volume (µL) in Millicell® Inserts

24-well, Greiner M8812

apical volume	0 (ALI)	100	200	300	400
basolateral volume	300	500	700	1000	1400

12-well, Greiner M8687

apical volume	0 (ALI)	200	400	600	800
basolateral volume	800	1000	1200	1400	1800

6-well, Greiner M8562

apical volume	0 (ALI)	1000	2000	3000	4000
basolateral volume	1000	1800	2800	3600	4200

↑ Recommended Volume

Standard Electrode Probe

Sanitizing

The Millicell® ERS 3.0 standard electrode probe is supplied non-sterile. To sanitize/disinfect, use a low temperature chemical sanitizing/disinfection solution such as 70% ethanol, 70% isopropanol, or enzymatic cleaners such as Tergazyme® enzyme detergent, Enzol® enzymatic detergent, or Alconox® enzymatic detergent. Bleach and 3% hydrogen peroxide are also compatible.

Caution: Ensure that only the electrode tip is submerged in liquid. Do NOT permit the solution to touch the upper probe housing and do not submerge the entire electrode probe in liquid.

Probe Placement

The Standard Electrode Probe fits 6.5-24 mm Millicell® hanging inserts and receiver plates (6-well, 12-well, and 24-well), and can be adjusted so the electrode reaches the bottom of the well plate while the adjustable ring rests on top of the well.

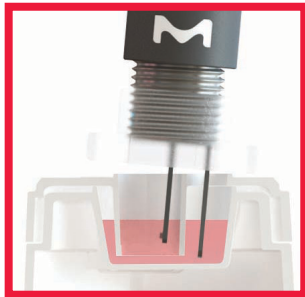
The electrode probe is designed to facilitate measurements of the membrane voltage and resistance of cultured epithelia in tissue culture wells. The length of the electrode tips allow the longer (basolateral) electrode to touch the bottom of the well containing the membrane insert, while preventing the shorter (apical) electrode from reaching the bottom of the membrane insert. This feature ensures proper positioning between the electrode and the cell layer in the cup during the trans-membrane measurement.

1. Make sure the volume of liquid in both apical and basolateral chambers is high enough to cover the entire electrode for most stable readings.
2. Place the electrode in the sample insert/well so the basolateral tip almost touch the bottom of the wells without probe angling on top.

Note: The apical (shorter) leg of the multiwell electrode probe does not touch the membrane surface by design and is at the optimal depth.

Standard Electrode Probe (6-, 12-, 24-well compatible)

Electrode probe is **not properly** placed



Electrode probe is **correctly** placed



Instrument Software Navigation

Main Menu Options

Work Offline

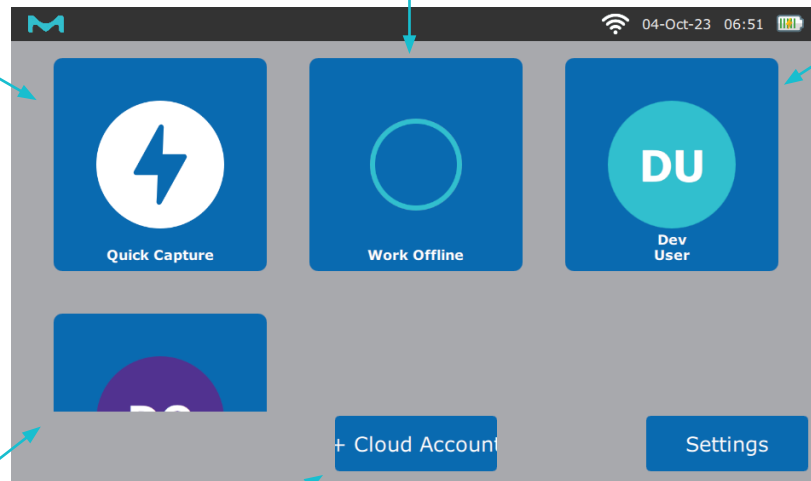
Used to create plate maps of experiments and record data specific to a plate's layout/grouping. Data will be stored on the instrument and users will have an option to export all data related to a plate map to a USB drive.

Quick Capture

The fastest way to begin reading TEER or TEPD. It will record values numerically with the option to export results to USB.

Caution: The data will not be saved in quick capture mode, unless a USB is used.

User profiles tiles are shown with the user's initials



Millicell® Cloud User

Required to create plate maps of experiments and record data specific to a plate's layout/grouping. Data will be stored in the Millicell® Cloud and users will have the option to view and export their data from the cloud website.

<https://app.millicellcloud.com>

+ Cloud Account

Adds a Millicell® Cloud account to this instrument.* Click this option, input the user's email address and password and press the Login button.

Settings

contains:

- Date & Time options
- Wireless Settings
- Check Verification
- Software Update
- About

Accessing through wireless network, users can:

- When adding an account, the user has the option to setup a pin. The pin can be changed by clicking on Settings in the user profile.
- When removing an account from this device, press and hold the user profile icon until the delete button appears. Click DELETE to confirm. This will NOT remove their updated data from the cloud.

Quick Capture Mode

(offline testing without plate maps)

Quick Capture Mode saves measurements sequentially on the left side of the screen. The top right side shows the well reading, temperature, and raw ohm or mV value.

Button Functions:

- **Tare Buffer** removes the current value from readings taken after it. Effectively performing background subtraction during live readings.
- **Volt Mode** will switch reading modes from TEER (resistance/ohm) measurements to voltage (mV) measurements. Volt Mode will change from TEER measurements to TEPD measurements.
- **Save & Next** records the onscreen well value, raw value, and temperature for the user.

Note: The footswitch accessory will serve the exact purpose as this button; pressing the footswitch will yield the same result as pressing this button.

- **Previous Well** to re-read a well and avoid a double measurement of the same well. This will erase the most recent measurement.

- **Finish** after the desired measurements for this experiment/plate is completed. After pushing Finish in Quick Start mode, instrument will prompt the user to save the results to a USB.

Note: If the user ignores the save to USB prompt, the data will not be saved.

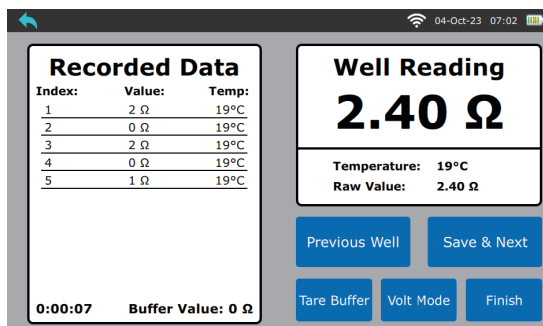
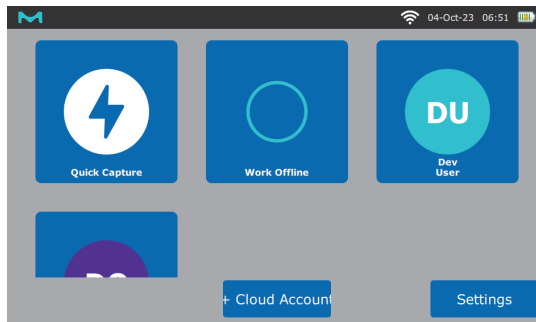


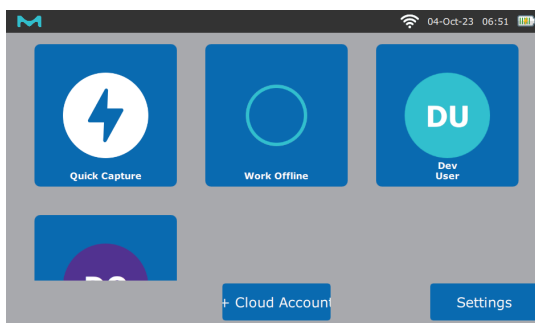
Plate Setup and Creating a New Plate

The Millicell® ERS 3.0 Digital Voltohmmeter works best when data is stored in an experimental plate, which will append the measurement data with the user inputted meta data assigned to grouped wells within a plate.

From the main menu, users can find their saved plates under their user Millicell® Cloud account. If a user does not have a Millicell® Cloud account, the device will locally store all plates in the Work Offline folder.

1. Click on the user profile/Work Offline folder.

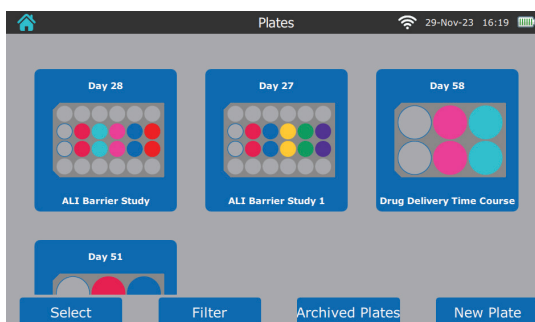
Note: If your lab has more than one Millicell® ERS 3.0 Digital Voltohmmeter in the same organization, data can be accessed through your cloud account from the other device. <https://app.millicellcloud.com>



2. Click on the New Plate button.

Note: From this screen you can also:

- Select an existing experimental plate map to continue an experiment.
- Select plates for archiving/deleting
- Filter active plates (by user, project, cell lines)
- View Archived Plates (finished experiments)

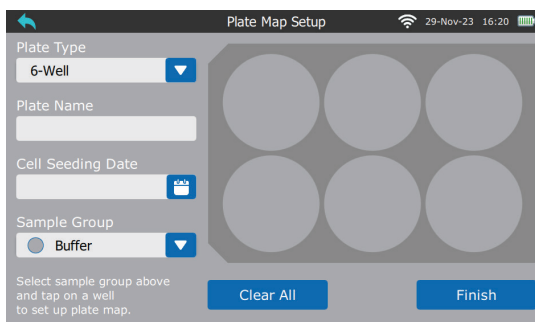


3. To setup the experimental plate, complete fields on the Plate Map Setup screen. The Plate Name is required to set it aside from other plates in this profile.

Note: The back button at the top left of the screen will bring you back to the user profile menu WITHOUT saving the plate. To save the plate click Finish.

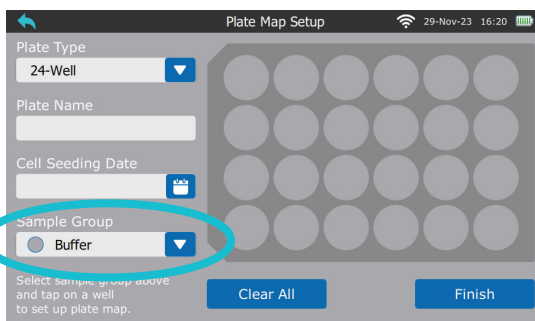
4. Click Plate Type (top left corner of the screen) and select the plate size from the drop-down menu.

Note: The plate image on screen will change to the plate type selected.

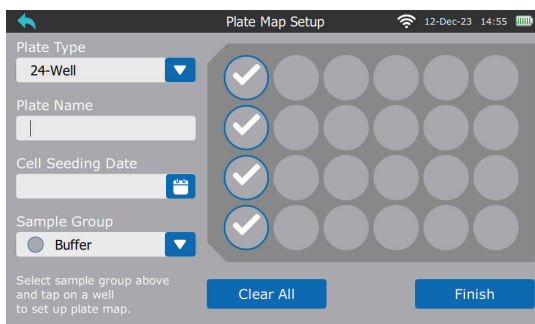


5. On the left side of the screen click the sample groups field to generate the drop down menu. Every plate the user makes will begin with a Buffer Only group; this group tells the instrument that these are the wells that get averaged and subtracted from sample wells as part of the background well/buffer subtraction.

Note: The user can assign their choice number of wells to this group by selecting the group in the drop-down menu, and then by pressing on the wells on the visible plate map. If wells are not selected they will be skipped during measurement taking.

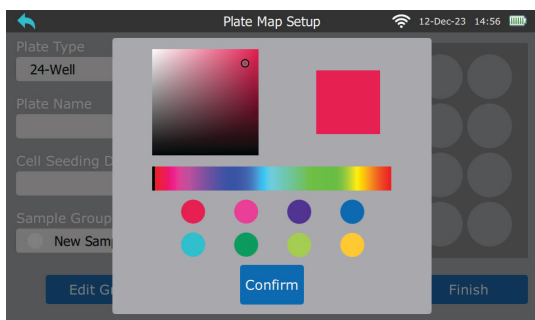
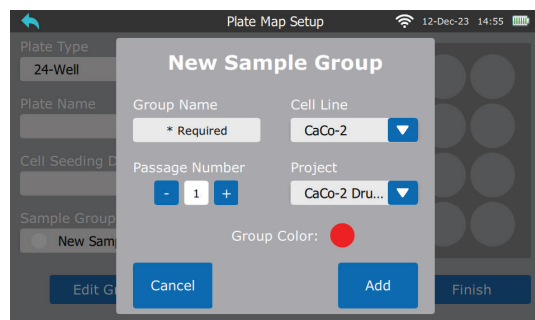


- All wells should be assigned to groups for the instrument to recognize them for measurements. Select Sample Group and select New Sample Group from the drop down menu.



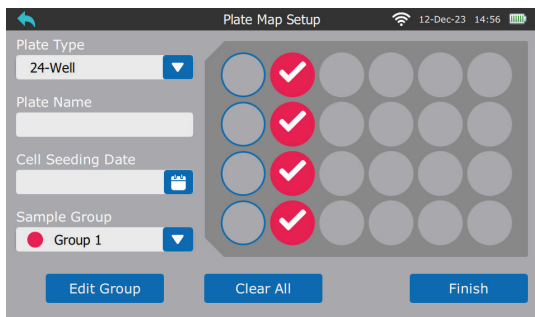
- The New Sample Group menu will pop up. Enter group name, cell line, passage number, project name, and group color (this color will be displayed on your plate map).

Note: Information on the instrument cannot be edited after pressing FINISH.

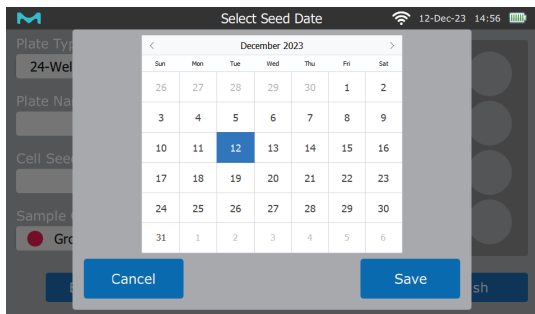


- To add the next group to the plate map, select the group in the drop-down menu and the appropriate wells on the plate.

Note: Wells can be unassigned and reassigned to different groups by selecting them again with a different group selected in the Sample Group drop-down.

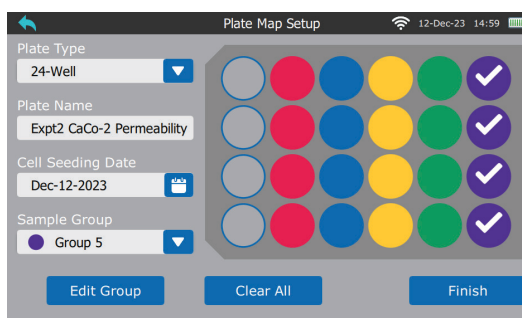


- When the Millicell® ERS 3.0 Digital Voltohmmeter is connected to the internet, the date and time may need to be manually updated. This is especially helpful in populating the Seed Date (the date the cells are added to the membrane inserts in the plate).



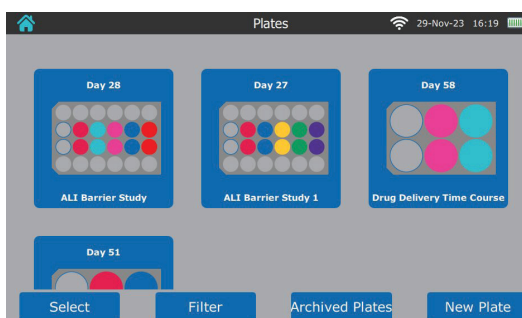
- After adding groups to the plate, select Finish to save the plate and return to the user profile menu.

Note: Do NOT select the Clear All or back arrow buttons. This will erase the plate you have just created without saving it.



- The plate will now appear in the user profile chronologically with the name the user assigned to the plate visible underneath the plate map image.

Note: To free up space in this menu, mark plates as archived.



Archiving a Plate

- Select the Experimental Plate map you would like to archive.
- At the bottom of the Experimental Plate map screen, select Archive. Archiving hides the plate and marks it as inactive (measurements and data cannot be added to this plate).

Note: Archiving is reversible if needed.



Measuring TEER

(Trans-Epithelial Electrical Resistance)

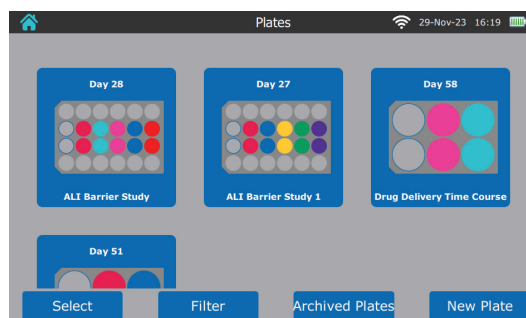
1. Remove the plate of cells from the incubator and allow them to reach room temperature before starting measurements.

Note: Resistance measurements are a function dependent on temperature, as temperature decreases, the resistance will increase. Stable temperature is the best condition for taking measurements.



2. From the user profile screen, select which Plate to add data.

Note: If there is no plate created for this experiment, please follow the instructions above to add a plate or select the home button and follow instructions for [Quick Capture Mode on page 10](#). See [Plate Setup and Creating a New Plate on page 11](#).



3. After selecting your plate, verify that the plate is correct before proceeding. The Back button will return the user to the user profile menu.

4. From a selected plate,

- **Past data** will display the values from the previous measurements in the well the measurement was taken. View by date & time and measurement type.

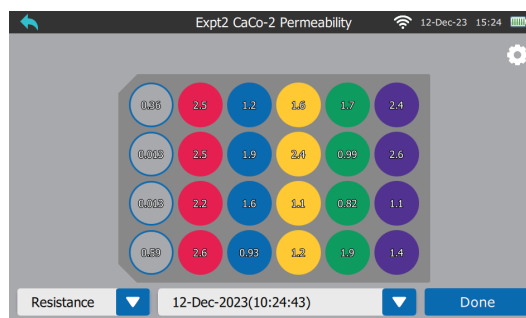
- **Export** will export all the data associated with the selected plate to the USB device.

Note: The Export button is only present in Work Offline mode. To retrieve data while measuring in a Millicell® Cloud Account, go to the Millicell® Beta Cloud to view and download measurements.

<https://app.millicellcloud.com>

- **Archive** to designate the plate as no longer active. It will keep previously recorded data but will not be able to add new data.

- **Add Data** to take additional measurements for TEER, TEPD, and Temperature.



- Select the Sample Group Order to take measurements.

Adjust the order by using the blue triangles on the sides of the group names. The groups will be read in top down order; up triangles will make the group read sooner and down triangles will put the group later.

Alternatively, the Ignore Groups option will give users the ability to read a plate directionally instead of by group. Users can select to read by row, column, or snaking patterns.

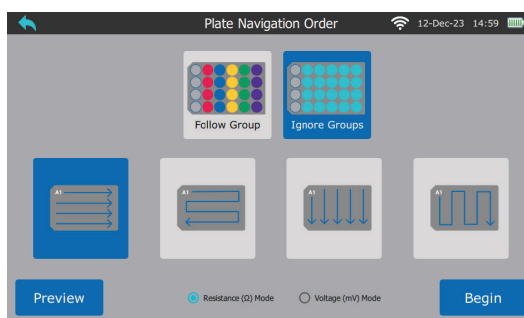


- Select Resistance (Ω) Mode or Voltage (mV) Mode

The Preview button will display the order the wells will be read in, based on the selections on this screen.

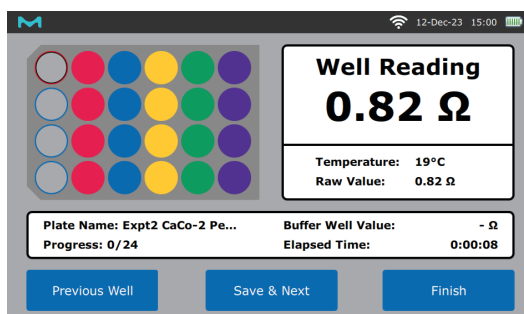
- Select Begin to start reading.

Caution: When moving probe electrodes from one sample well insert to another, it is best NOT to rinse the electrodes with Milli-Q[®] water or distilled water. Electrodes on the probe may take several minutes to recover from exposure, during which time the measurement may drift by a few millivolts or ohms. If it is necessary to wash the electrodes between measurements to avoid carryover of one sample into the next, the electrodes should be rinsed with the experimental culture media.



- The Measurement screen shows the plate layout, the Plate Name, the user progress for measuring wells, Buffer Average (if applicable), and the time since measurements started on the left side. On the right side of the screen is the Well Reading, which will differ from the raw value once the background wells have been read. The other visible outputs are the well Temperature and the Raw Value of the well. To record a measurement select Save & Next as described in [Quick Capture Mode on page 10](#).

Note: The instrument highlights the well that it thinks is actively being read in a red circle on the plate map, if the user would like to read a different well, the user can select the plate map and then chose another well manually.



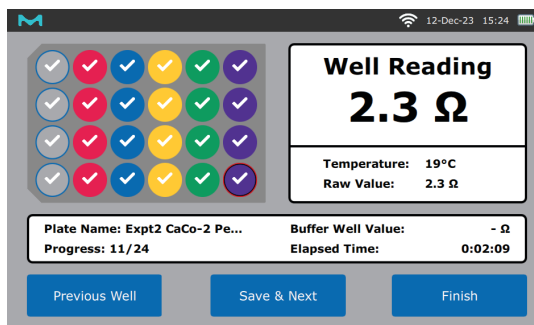
- Selecting the well map to expand the plate into a full screen view. Select one of the wells to read that well next or reread a well that has been already measured. This view also displays the measurement values for each well and select wells to reread.

Warning: Only one measurement per well is recorded at a time. Rereading a well in the middle of a plate will overwrite the first reading.

Note: To go back to the measurement screen, either select a new well on the plate map or select Cancel to return to the most recent well.



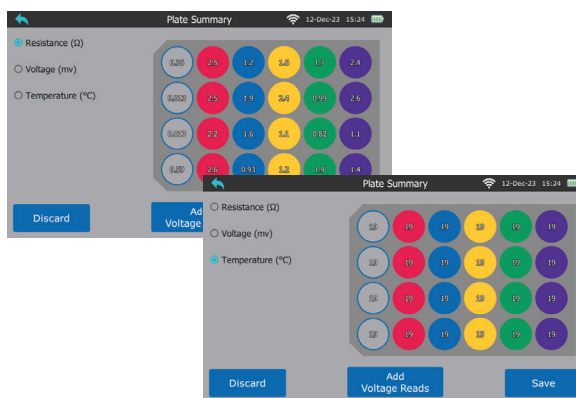
- Continue to take measurements by selecting Save & Next until all the wells have been read or the user has read their choice wells. When finished reading, select Finish.



- The next screen shows summary of measurements with toggle buttons for each measurement type: Resistance, Voltage, and Temperature.

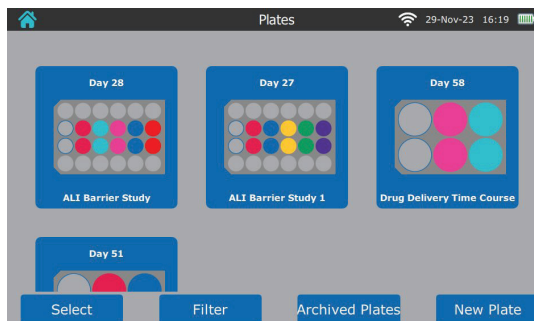
- If measurements are completed, select Save to save the data to the device or Millicell® Cloud account.
- To return to the measurement screen, select the back arrow at the top left of the screen.
- To restart the measurements in voltage mode, select the Voltage Mode button.

Note: Selecting Discard will delete this data from the device.



All changes to the data must be made before saving data (step 12).

- After selecting Save on the Plate Summary screen, the Millicell® Cloud User profile will display the active experiment plates.



Measuring TEPD

(Trans-Epithelial Potential Difference)

1. For optimal electrode probe TEPD measurements, leave the probe in the measurement solution for 15-30 minutes. This allows the buildup of ions to equilibrate between the electrodes. Take a measurement of the solution to verify that the reading is stable.

Note: The user might see the measurement slowly increase or decrease when taking readings in voltage mode, this is indicative of ion buildup on one of the electrodes and more soaking is necessary.



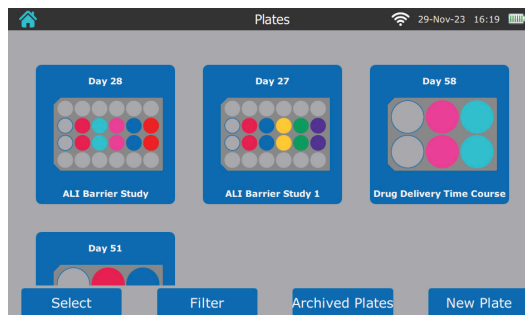
2. Remove the plate of cells from the incubator and allow them to reach room temperature before starting measurements.

Note: Stable temperature is the best condition for taking measurements.



3. From the user profile screen, select which Plate to add data.

Note: If there is no plate created for this experiment, please follow the instructions above to add a plate (see [Plate Setup and Creating a New Plate on page 11](#)) or select the home button and follow instructions for [Quick Capture Mode on page 10](#).

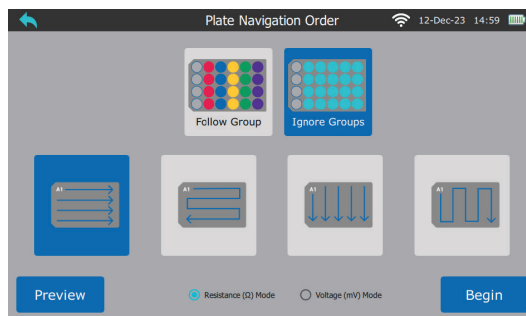
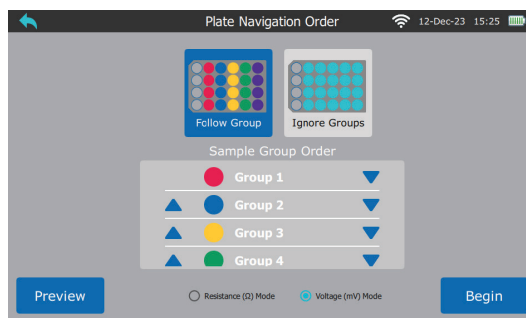


4. After selecting a plate, verify that the plate is correct before proceeding.
 - Select Add Data to take additional measurements for TEER, TEPD, and Temperature.
 - The Back button will return the user to the user profile menu.



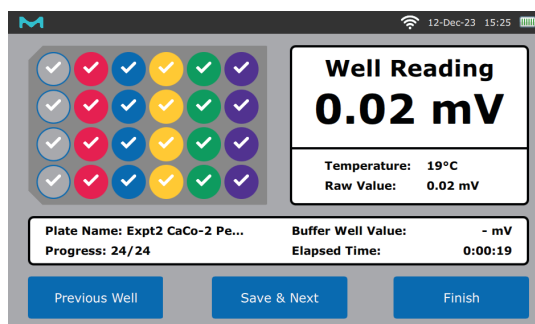
5. Select the Sample Group Order for taking measurements.
6. Select taking measurements Voltage (mV) Mode.
The Preview button will display the order the wells will be read in, based on the selections on this screen.
7. Select Begin to begin reading.

Caution: When moving probe electrodes from one sample well insert to another, it is best **NOT** to rinse the electrodes with Milli-Q® water or distilled water. Electrodes on the probe may take several minutes to recover from exposure, during which time the measurement may drift by a few millivolts or ohms. If it is necessary to wash the electrodes between measurements to avoid carryover of one sample into the next, the electrodes should be rinsed with the experimental culture media.



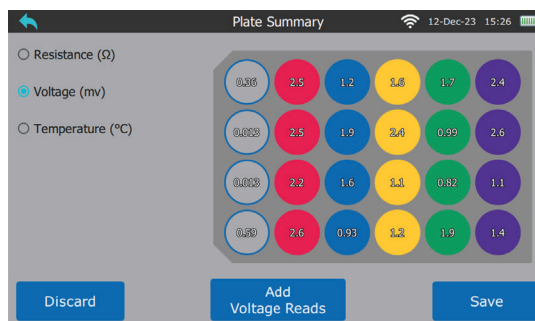
8. Measure the background wells. This is essential to calculating the true voltage measurement of the cells.
9. Select Save & Next to record measurements and read the entire plate.
10. Select Finish and continue to the Plate Summary screen.

Note: Check for ion drift by re-reading the first well at the end of a series of voltage measurements. This will inform the user if the value has significantly changed.

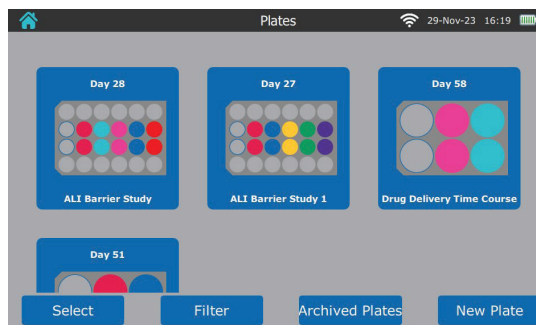


11. After selecting the Finish button, the screen will display the summary of measurements with toggle buttons for each measurement type: Resistance, Voltage, and Temperature.
 - If measurements are complete, select the Save button to save the data to the device and Millicell® Cloud account.
 - To return to the measurement screen, select the back arrow at the top left of the screen.
 - To restart the measurements in resistance mode, select the Resistance Mode button.

Note: Selecting Discard will delete this data from the device.



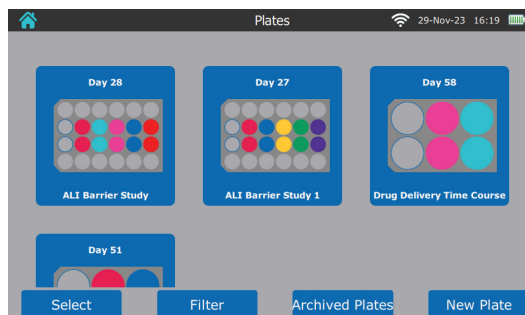
12. After selecting Save on the Plate Summary screen, the Millicell® Cloud User profile will be displayed with all of the active experiment plates shown.
13. Press individual wells to see the date/time with measurements or select Past Data to see the measurements displayed across all wells at a given date/time.
Selecting the back arrow or the Back button to return to their profile menu.



USB Data Exportation

(Work Offline mode only)

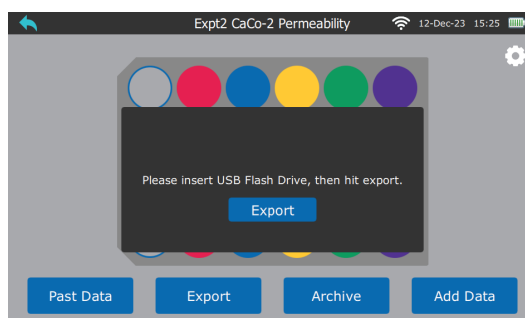
1. From the Work Offline user profile plate menu, select a plate that has had data collected.



2. In the Plate Menu, select the Export to export the existing data to a USB drive.



3. The instrument will prompt the user to insert their USB Flash Drive and then select Export to send data to their USB.
Note: The Millicell® ERS 3.0 requires USB Flash Drives to be formatted into FAT32 format or the USB will not be recognized or compatible.



Data Export to Cloud

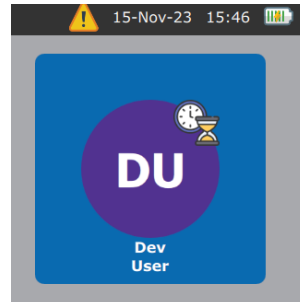
(cloud account)

After taking measurements, select Save. The data will automatically export over your network connection (Wireless network or ethernet) to the Millicell® cloud.

Note: The symbols pictured here indicates the stage of data export the instrument is undergoing. The Millicell® ERS 3.0 Digital Voltohmmeter is designed to save data temporarily on device when network connection is broken so that the data is not lost.

If the dongle is removed during measurements, the data will not upload to the Millicell® Cloud. A yellow triangle with an exclamation point will displayed to alert the user. In addition, the account associated with the failed upload will be marked with a symbol.

The next time internet connection is established, the data will automatically upload. The USB Wireless network dongle can be plugged into any of the instrument's three USB ports. It can stay plugged in during storage, usage, and charging.



Network Connection

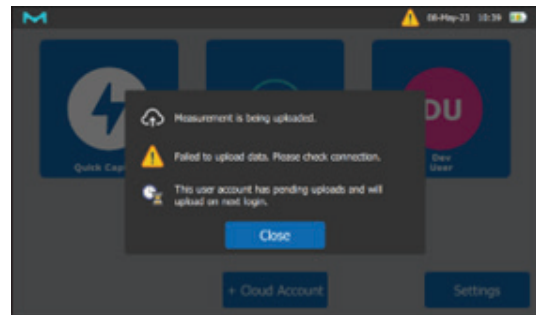
Using Ethernet

Connect the Ethernet cable (not provided) to the port on the back panel of the Millicell® ERS 3.0 Digital Voltohmmeter, and the other end to a working Ethernet outlet.



Using Wireless network

1. Plug the external USB Wireless network dongle (included) into a USB port on the back panel of the Millicell® ERS 3.0 Digital Voltohmmeter.
2. From the Home Screen, choose Settings.
3. Click Wireless Settings.
Note: You may need to contact your local IT administrator regarding your company or institution's network connection policies and guidelines.
4. Choose Network. The Millicell® ERS 3.0 Digital Voltohmmeter supports 2.4 GHz and 5 GHz networks.
5. Enter the Wireless network password and click Connect.



System Verification

The Millicell® ERS 3.0 Digital Voltohmmeter and electrode probe should be tested periodically to ensure proper operation. Test the full system when you first receive it, at the beginning of a series of measurements, and as needed during the course of the measurements to confirm stability.

Prior to each use, the following checks should be performed:

- [Testing with Verification Adapter](#) Instrument functionality check for resistance and voltage measurements.
- [Probe Verification](#) Electrode probe functionality check for resistance in KCl.

Testing with Verification Adapter

The verification adapter is an essential part of the Millicell® ERS 3.0 because it informs the user that their device is measuring Resistance and Temperature within appropriate ranges. A failure to read the verification adapter values within acceptable ranges is indicative of something wrong with the instrument or software.



1. Insert the verification adapter device into the instrument port labeled "Sensor" as seen in the picture on the left.

Note: The Mini-DIN Plug pins need to be correctly aligned with the port so that neither is damaged.

The Verification adapter can be used whenever a measurement is being taken, as long as the instrument is measuring in resistance mode.



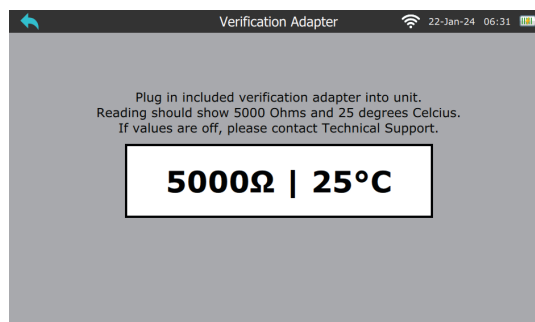
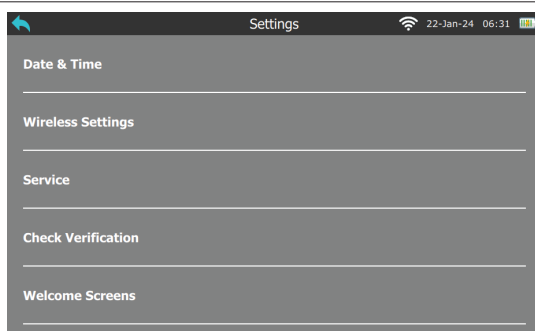
2. Open settings from the home screen menu.

3. In the settings menu there is an option to Check Verification. Open this option to check the verification with the verification adapter.

4. Verify the verification adapter reads within expected ranges. Back out to the main menu, and then unplug the adapter. Now you are ready to plug in the Electrode Probe for normal measurement taking.

Note: Acceptable ranges for the verification adapter are $5,000 \Omega \pm 5 \Omega$ and $25 \text{ }^\circ\text{C} \pm 1 \text{ }^\circ\text{C}$.

If the instrument is out of spec contact SigmaAldrich.com/techservice for recalibration.



Probe Verification

KCl testing solutions have stable resistance values for probe validation/verification. To make 160 mM KCl testing solution add 1.192 grams KCl to 100 mL of distilled water. (1M KCl = 74.54 g/M in 1 L DI water).

Use	Add DI H ₂ O	Results	Resistance Range
1.192 gm KCl powder	100 mL	100 mL of 160 mM KCl	14 ± 6 Ω
50 mL of 160 mM KCl	50 mL	100 mL of 80 mM KCl	32 ± 12 Ω
50 mL of 80 mM KCl	50 mL	100 mL of 40 mM KCl	52 ± 14 Ω
50 mL of 40 mM KCl	50 mL	100 mL of 20 mM KCl	98 ± 32 Ω
50 mL of 20 mM KCl	25 mL	50 mL of 10 mM KCl	195 ± 66 Ω

At lower molarities (20 mM and below), a DMEM coated electrode will begin to show instabilities at the higher concentrations. If the resistance readings in the KCl mixtures are over 10% out of range, then the electrode may need cleaning. Maintaining the electrode and properly cleaning it are important to its function. An Excel scatter plot of the molarity vs. resistance (as a power) should show an R-squared value > 0.99. To get this result, graph your data as a scatterplot with molarity on the x-axis and resistance on the y-axis, add a trendline and in trendline options select Power and Display R-squared value on chart.

Note: These resistance readings can vary due to fluid volume and electrode depth. These values are a reference and are taken without membrane culture inserts present. If the R-squared value is acceptable, then the electrode can be used even if it is slightly out of range. If the resistance readings in the KCl mixtures are over 10% out of range, then the electrode may need cleaning. Buildup of media on the electrode will cause instabilities.

TEER Detection Method Example Application

The Millicell® ERS 3.0 Digital Voltohmmeter is designed for non-destructive testing for epithelial mono-layer confluence in cell cultures using low current and voltages. The low AC current produced avoids electrode metal deposits and adverse effects on tissues which can otherwise be caused by higher DC currents. Confluence of a cellular mono-layer is determined by an increase or a plateau in tissue resistance detected using the Millicell® ERS 3.0 Digital Voltohmmeter.

MDCK culture media

- Dulbecco's MEM with high glucose
- 10% FBS
- 1% (1x) NEAA
- 1% Penicillin/streptomycin
- 4 mM L-glutamine

Seeding Cells onto a 24-well Millicell® Membrane Insert

1. Culture MDCK cells until they are 60-80% confluent.
2. Aspirate media from the culture flask.
3. Add sterile PBS for 3 minutes at room temperature.
4. Aspirate liquid from the culture flask.
5. Add trypsin to the culture flask and incubate in a 37 °C, 5% CO₂ cell culture incubator.
6. Stop the trypsin reaction by adding equal parts complete cell culture media to the flask.
7. Collect liquid suspension and spin down in the centrifuge at 300 x g for 3 minutes.
8. Resuspend cells in 1 mL of cell complete culture media.
9. Count cells using trypan blue solution (T8154-100mL) and a Millicell® Disposable Hemocytometer (MDH-4N1). Mix 90% Trypan blue with 10% cell suspension (for example, 180 µL Trypan Blue solution and 20 µL cell suspension).
10. Count live cells on the hemocytometer and calculate cells per milliliter.
11. Add 900 µL of complete media to the basolateral side of the 24-well membranes.

12. Resuspend cell suspension in appropriate volumes to dispense 200 μ L of suspension containing the desired cell seeding density (e.g., 1,250 cells/insert, 2,500 cells/insert, 5,000 cells/insert).
13. Place in the 37 °C, 5% carbon dioxide cell culture incubator and change media every other day.

Note: Pre-warm complete cell culture media before adding to Millicell® hanging insert and receiver plate.

Measuring Cell Resistance

1. Allow plate to reach room temperature (15-30 minutes).
2. Load plate map and assign wells to groups on the Millicell® ERS 3.0 Digital Voltohmmeter as shown earlier in this manual in the pictured walk through titled: "Plate setup and creating a new plate map".
3. Test the Millicell® ERS 3.0 Digital Voltohmmeter using the verification adapter. Make sure that the resistance measures $5,000 \Omega \pm 5 \Omega$ and the temperature reads $25 \text{ }^\circ\text{C} \pm 1 \text{ }^\circ\text{C}$.
4. Immerse the electrode so that the shorter tip is in the apical side of the Millicell® Hanging Cell Culture Insert, and the longer tip is in the outer well. The shorter tip should not contact cells growing on the membrane and the longer tip should just touch the bottom of the outer well. To ensure stable and reproducible results, make sure that the adjustable ring rests flat on the top of the insert and the electrode is held steady at a 90 °C angle to the plate/insert.

Note: If rinsing is required between measurements to prevent sample carryover, use cell culture media rather than Milli-Q® water or distilled water.

5. Record the resistance by pressing save and next on screen.
6. Determine blank resistance by adding media or electrolyte solution to a blank well/cell culture insert (i.e., the cell culture insert without cells and complete cell culture media).
7. Measure the resistance across the blank insert and use this value as a "zero" or "background" resistance level. To obtain true tissue resistance, subtract the resistance reading across the blank insert from the resistance reading across the tissue (cell culture insert with cells).

The resistance is inversely proportional to the area of the tissue. The larger the membrane, the lower the resistance.

Unit Area Resistance ($\Omega \times \text{cm}^2$) = Resistance (Ω) \times Effective membrane Area (cm^2)

Note: Resistance can vary based on sample temperature, pH, and electrode depth in solution.

Maintenance and Storage

Instrument

Cleaning

Wipe the body of the Millicell® ERS 3.0 Digital Voltohmmeter with 70% ethanol. The instrument is NOT submersible.

CAUTION: To prevent corrosion, do not splash or spill saline solution or culture media on the instrument.

Storage

When not in use, the Millicell® ERS 3.0 Digital Voltohmmeter should be powered off and disconnected from the power cable, as continuous charging may shorten battery life. To prevent the digital LCD display from darkening, do not leave the instrument in direct sunlight for long periods of time. Store the system between 15-30 °C with 20-80% non-condensing relative humidity.

Battery

Normal Use

The battery level is indicated at the top right of the display screen. When the battery charge is low, the charge icon in the corner of the touch screen will turn red. To avoid automatic shut-off, plug the instrument to the power adapter.



The battery may be charged at any time in the discharge cycle and can be charged continuously without damage, using the supplied charger. Continuous charging may shorten battery life.

The instrument should hold a charge for approximately 4 hours of continuous use. The instrument screen will go to sleep after 15 minutes of inactivity. The instrument will shut off automatically when the battery runs low to avoid damage to the instrument.

Changing the Battery

The Millicell® ERS 3.0 Digital Voltohmmeter is equipped with a rechargeable Lithium-Ion battery pack.

CAUTION: To prevent battery damage, use ONLY the supplied charger.

1. Using a flathead screwdriver, unscrew the captive screw on the back panel of the Millicell® ERS 3.0 Digital Voltohmmeter and separate the panel from the base of the instrument.

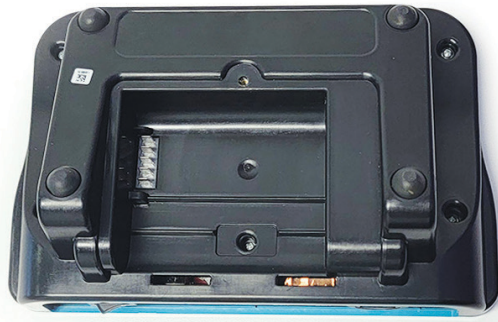


2. The battery pack is mounted to the base of the instrument. Remove the battery pack from the instrument by pulling up on the battery pack tab. This will decouple it from the instrument and lift the battery pack up.

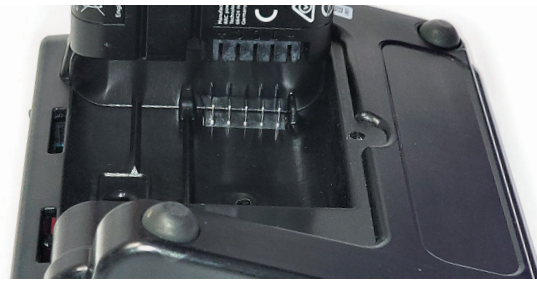


-
- Note the position of the battery pack mating connector. This connection type ensures that the battery pack can only be successfully connected in one orientation.

CAUTION: Connecting the battery pack connector improperly may result in personal or equipment damage.



-
- To connect a new battery pack to the instrument, line up the battery mating connector on the instrument to the corresponding battery pack connection so that the protruding touch points match (as shown). Push the connector straight down over the pins on the instrument. Do not force the connector; if aligned properly, it should slide into place with little effort.



-
- Reassemble and re-secure the back panel. Using a flathead screwdriver fasten the screw into place to secure the panel.
 - Charge new battery before use.
 - Dispose of old battery in accordance with local regulations.



Electrode

Cleaning

With repeated use, the electrode surface can become coated with protein, salts, or other foreign materials. This build-up can degrade the performance of the system. After every use, rinse the electrodes with Milli-Q® water and store them as indicated below.

Electrode Chemical Compatibility

Chemical Requirement	Length
Ethanol/isopropanol alcohol, 70%	15 minutes
Ethanol wipe, 90%	1 minute
Enzyme detergent, 1% (Tergazyme®, Ezcol®, or Alconox®)	See below*
Hydrogen peroxide, 3%	30 minutes
Sodium hypochlorite (bleach), 5%	15 minutes
Sodium hypochlorite (bleach), 10%	15 minutes

CAUTION: Repeated rubbing or scratching will damage the coating of the electrode, which can severely damage the integrity and accuracy of measurement taking.

Also Tested Safe to Use with Electrodes

Cell culture medium with FBS

Prolonged exposure to electrolyte solution (KCl/PBS)

Clean the Electrodes with Enzymatic Cleaner

1. Rinse the electrodes with Milli-Q® water or distilled water.
2. Make a 1% solution of enzymatic cleaner detergent and suspend the electrode tips in the solution with the exposed electrode surfaces fully immersed. If desired, gently brush the electrode surfaces with a soft brush (e.g., toothbrush).

*If the electrode is...	Soak Time
Not cleaned routinely	12 hours
Is cleaned on a weekly cleaning schedule	30-60 minutes
Is cleaned daily	5 minutes

3. Rinse well with Milli-Q® water and allow to air dry. See [Storage](#) below.

Storage

Sanitize electrodes with 70% ethanol and immediately rinse with Milli-Q® water or distilled water. Do not allow electrode to dry without first rinsing to remove salts and proteins.

After proper cleaning, store electrodes protected from light. Sunlight and UV light from cell culture hoods may damage the electrode and probe. Protect electrodes from scratching and physical damage.

Millicell® Cloud Account

Subscribing to the Millicell® Cloud account optimizes your workflow efficiency and enables seamless data collaboration. To subscribe and set up the account for your organization, a primary contact email address must be established. One organization account can accommodate multiple Millicell® ERS 3.0 Digital Voltohmmeters as well as other Millicell® Cloud compatible devices. After the organization account is established, individual user accounts can be invited to join to collaborate on data.

<https://app.millicellcloud.com>

Once you are logged into your Millicell® Cloud organization, you can navigate to the following sections:

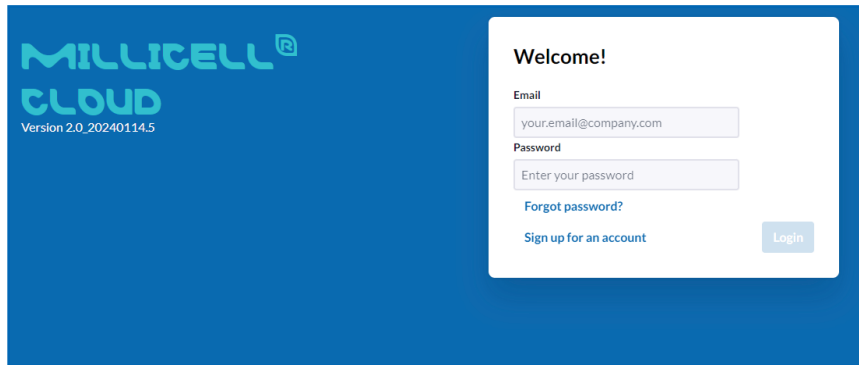
Projects	Create & manage projects
Measurements	Create & manage plates
Trends	View charts & plots about your data
Account	Manage your account
Organization	Manage organization and invite others to join your Millicell® Cloud organization
Cell Line	Filter based on user cell lines, store information about each measurement set

For the best experience we recommend using the latest versions of Google Chrome™, Microsoft Edge®, or Safari® browsers.

Establish the organization account

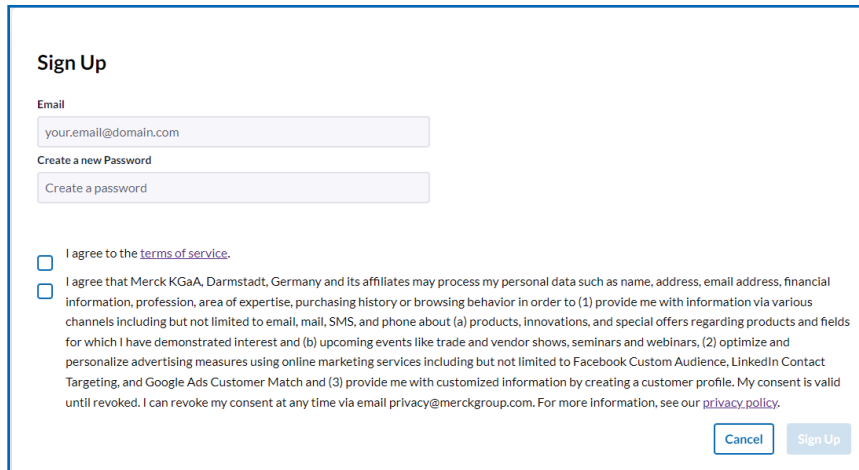
1. The primary contact email address will receive a Welcome email containing a link to create the Millicell® Cloud account for your organization.

Note: If you did not receive an email, someone else may be setting up your organization.



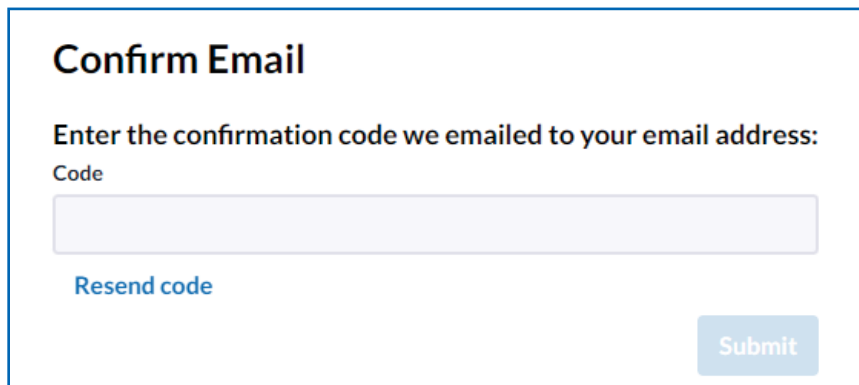
The image shows the Millicell Cloud login interface. On the left, the Millicell Cloud logo is displayed with the version number 2.0_20240114.5. On the right, a white box titled 'Welcome!' contains a login form with fields for 'Email' (pre-filled with 'your.email@company.com') and 'Password' (with the placeholder 'Enter your password'). Below the password field are links for 'Forgot password?' and 'Sign up for an account', and a 'Login' button.

2. Click sign up for an account. Add the email address and password. Read and accept the Terms of Service and Privacy Policy. Click sign up.



The image shows the 'Sign Up' form. It includes fields for 'Email' (pre-filled with 'your.email@domain.com') and 'Create a new Password' (with the placeholder 'Create a password'). Below the fields are two checkboxes for terms of service and privacy policy. The first checkbox is for 'I agree to the [terms of service](#)'. The second checkbox is for a detailed consent statement: 'I agree that Merck KGaA, Darmstadt, Germany and its affiliates may process my personal data such as name, address, email address, financial information, profession, area of expertise, purchasing history or browsing behavior in order to (1) provide me with information via various channels including but not limited to email, mail, SMS, and phone about (a) products, innovations, and special offers regarding products and fields for which I have demonstrated interest and (b) upcoming events like trade and vendor shows, seminars and webinars, (2) optimize and personalize advertising measures using online marketing services including but not limited to Facebook Custom Audience, LinkedIn Contact Targeting, and Google Ads Customer Match and (3) provide me with customized information by creating a customer profile. My consent is valid until I revoke it. I can revoke my consent at any time via email privacy@merckgroup.com. For more information, see our [privacy policy](#).' At the bottom right are 'Cancel' and 'Sign Up' buttons.

3. A verification code will be emailed to the person purchasing the system. Enter the verification code and click submit. Click log in again and sign in to complete the process. If you do not receive a code, click the resend code link or see [Troubleshooting on page 32](#).



The image shows the 'Confirm Email' form. It features the heading 'Confirm Email' and the instruction 'Enter the confirmation code we emailed to your email address:'. Below this is a 'Code' field with a large text input box. A 'Resend code' link is positioned below the input box. At the bottom right is a 'Submit' button.

- Click "I received an access code and want to set up an organization".

Welcome to your Millicell® Cloud account!

Before you can start saving images to the cloud, you need to create or join an organization. Organizations allow you to collaborate and share data with colleagues.

I received an access code or have a free trial code and want to set up an organization.

Someone else will set up the organization for my team.

I want to delete my account

Note: Once your organization's account is established, the organization owner can invite individual to the organization account.

- Complete the form, entering the access code from in the email. Click create to complete the organization setup process.

Note: The free trial code offer (see bottom of device) can provide an initial access code.

Create Organization

Fill in the form below to create your Organization.

Your First Name

Your Last Name

Organization Account Name

Your organization homepage will be:

✖ This Section is required.

Access Code

OR

Free Trial Code

Serial Number of Device with Free Trial Code

Cloud Usage and Login

An email will be sent containing your login and password. This login and password combination will be used on the instrument to add cloud account and online to access your data. The data will only be uploaded to the Millicell® Cloud when the device has internet connection (Wireless network or ethernet). If you cannot connect to the internet on your device, then you will not be able to send data to the cloud or use the cloud user account on device. The work offline option will always be accessible regardless of network connection.

Downloading Data

1. Open a link to the cloud <https://app.millicellcloud.com> and use the user login supplied via email.
2. Click the link to the Millicell® Cloud and input the provided username and password credentials.
3. After logging in, you will be faced with the Measurements page. This page will display all your active experiments separated by plate.
4. Select the plate containing the data of interest. This should open up an information page with your plate map, calculations, and information related to the plate.
5. Click on any well(s) of the plate map to open an accordion menu displaying data.
6. To download a CSV file of all of the raw data, scroll down to the bottom of the page and select Download Plate Data (CSV).
7. This will download your data to your default downloads folder on your device.

Troubleshooting

Problem	Solution
Instrument/Cell Culture	
Instrument is frozen or not working	Turn the instrument off and back on again. WARNING: unsaved data will be lost.
I lost the website for the Millicell® Cloud and cannot get to my data.	https://app.millicellcloud.com
Cell death/yellowing media	Change media every other day to ensure cells have enough nutrients and maintain vital cell signaling for monolayer/tight junction formation. Check for bacterial contamination in user culture. Use antibiotics preventatively. Be careful not to touch or poke the membrane while aspirating or taking measurements.
TEER measurements were high, but follow up assays failed	Do not leave cells outside of the incubator for prolonged periods of time before assays. This can negatively impact Lucifer yellow permeability assay results as well as other similar barrier assays.
Unstable resistance or voltage reading, (voltage drift) during use	Make sure the electrodes on the TEER probe are fully submerged in media for most stable readings. The Millicell® ERS 3.0 probe should be able to stand hands free for measurements. Make sure there is no vibration from the cell culture hood or user hand tremors. Hold the TEER probe at the same depth in each well/insert for consistent and comparable readings. To ensure stable and reproducible results, make sure that the electrode is held steady and at a 90° angle to the plate insert. Resistance can vary with temperature. TEER readings increase (higher ohms Ω) at lower temperatures (lower °C). Be sure to wait for samples to acclimate to room temperature before taking readings. Resistance is affected by media pH. Higher carbon dioxide content will be present in samples that are pulled immediately out of an incubator. This will automatically balance with the atmospheric carbon dioxide while acclimating to room temperature. Clean TEER electrodes periodically with an enzymatic detergent such as Tergazyme®, Enzol®, or Alconox® to eliminate protein buildup on the electrode from cell culture media. If rinsing is required between measurements to prevent sample carryover, use cell culture media rather than Milli-Q® water or distilled water. TEPD or voltage mode readings may require the probe to equilibrate in measurement solution for 15-30 minutes without a membrane separating the two electrode heads. TEPD/Voltage mode works better on battery only. Use the verification adapter to verify that the instrument is measuring properly. The instrument is working correctly when the verification adapter reads 5,000 Ω ± 5 Ω and temperature at 25 °C ± 1 °C.
The instrument does not detect out of solution between well measurements and does not take new temperature measure	Wait between wells with the probe in the air until the screen displays three dashed lines to indicate out of solution.
Instrument will not power on when disconnected from charge cable or meter runs briefly, then powers off	Batteries are discharged or insufficiently charged. Use the instrument with power cable attached.

Problem	Solution
Instrument battery will not charge when connected to charger	Insecure connection at the power jack on instrument. Defective power cable. Contact Technical Service at SigmaAldrich.com/TechService .
Intermittent instrument reading	Loose or broken electrode connector or cable. Replace the electrode. Malfunctioning system. Contact Technical Service at SigmaAldrich.com/TechService .

Network Errors

Unable to find desired wireless network	The Millicell® ERS 3.0 Digital Voltohmmeter supports 2.4 Ghz-5 Ghz wireless network when used with the USB wireless network dongle (included). If the issue persists, try moving the instrument closer to the desired Wireless network router or use the Ethernet port.
Cannot connect to wireless network	Wireless network policies or guidelines may have been put in place by your company or institution to govern the use of networks. Please contact your local IT department to learn what these requirements are. It may be possible to safe list (whitelist) this device by giving them the MAC address found in wireless internet settings.
Unable to mount USB flash Drive	Only a Standard Universal Serial Bus (USB) Type A connector with FAT32 formatting will be recognized. Using cloud storage is recommended.
User login timing out	Try moving the Millicell® ERS 3.0 closer to the desired Wireless network router or plug the Millicell® ERS 3.0 into an Ethernet port then log back into the user account. If problem persists, it is possible the network security infrastructure is blocking network traffic, please contact your Network/System Administrator for more assistance.
No USB flash drive found Occurs when USB flash drive is plugged into the device	Only a Standard Universal Serial Bus (USB) Type A connector with FAT32 formatting will be recognized. Ensure the drive is fully plugged into the USB port on the back device, wait a few seconds, and then try again. Using cloud storage is recommended.
No update file found Occurs when updating software from a USB flash drive	Ensure the USB is properly formatted FAT32, plug in to the back of the instrument, wait several seconds, and then try again.
Update file corrupt or invalid Occurs when updating software from a USB flash drive	Try deleting the update file from your computer and the USB drive. Re-download the update file and make sure the download completes before copying the file over to the USB drive. Do not change the name of the update file or add any numbers or symbols to the filename.
Unable to update Occurs after updating software from a USB flash drive	Try deleting the update from your computer and the USB drive. Re-download the update file and make sure the download completes before copying the file over to the USB drive.

Data Workflow

Did not receive Millicell® Cloud invitation email	Contact Technical Service at SigmaAldrich.com/TechService .
Did not receive verification email	Contact Technical Service at SigmaAldrich.com/TechService .
Instrument is frozen or not working	Turn the instrument off and back on again. WARNING: Unsaved data will be lost.

Product Ordering

Purchase products online at SigmaAldrich.com.

Millicell® ERS 3.0 Digital Voltohmmeter

System and Components



Device	Catalogue Number
Millicell® ERS 3.0 Digital Voltohmmeter with instrument, standard electrode, power supply, dongle, ver. device, SW	MERS03000
Millicell® ERS 3.0 Standard Adjustable Electrode (for 6-, 12-, 24-well plates)	MERS03SAP
Millicell® ERS 3.0 Verification Device	MERS03VER
Battery for Millicell® ERS 3.0	MERS03BAT
Foot Pedal for Millicell® ERS 3.0	MERS03PED
Millicell® ERS 3.0 Wireless Network USB Dongle	MERS03USBD0N
Millicell® Cloud Free Trial Software Subscription	MERS03CLTRIAL
Millicell® Cloud 1 Year Software Subscription	MERS03CL1YR
Millicell® Cloud 5 Year Software Subscription	MERS03CL5YR
Millicell® Cloud 10 Year Software Subscription	MERS03CL10YR

Related Devices and Accessories



Device	Catalogue Number
Millicell® Digital Cell Imager	MDCI10000
Millicell® DCI Wireless network USB Adapter Compatible with Millicell® ERS 3.0 Digital Voltohmmeter	MDCI1USBD0N
Millicell® DCI Power Supply Compatible with Millicell® ERS 3.0 Digital Voltohmmeter	MDCI1PWRSUP
Scepter® 3.0 Handheld Automated Cell Counter Kit, includes pkg of 40 µm Scepter 3.0 sensors	PHCC340KIT
Scepter® 3.0 Handheld Automated Cell Counter Kit, includes pkg of 60 µm Scepter 3.0 sensors	PHCC360KIT
Trypan Blue Solution, 100 mL	T8154-100mL
Millicell® Disposable Hemocytometer, 50 pk	MDH-4N1-50PK

Cell Culture Plates

Greiner Bio-One 100 ea M8812-100EA
CELLSTAR® Cell Culture
Multiwell 24-well plates
with lids

Greiner Bio-One 100 ea M8687-100EA
CELLSTAR® Cell Culture
Multiwell 12-well plates
with lids

Greiner Bio-One 100 ea M8562-100EA
CELLSTAR® Cell Culture
Multiwell 6-well plates
with lids

Millicell® 96-well Cell Culture Insert Plates

Single-well Feeder Tray	Multiwell 96-well Receiver Plate	Membrane Pore Size	Qty/ PK	Catalogue Number
yes	no	PCF 0.4 µm	5	PSHT004R5
no	yes	PCF 0.4 µm	5	PSHT004S5
yes	yes	PCF 0.4 µm	1	PSHT004R1
yes	yes	PET 1.0 µm	1	PSRP004R1
yes	no	PET 1.0 µm	5	PSRP004R5
Trays for 96-well receiver plates with lids			5	MACAC0RS5

Millicell® Hanging Cell Culture Inserts

Pore Size	Plate Size	Qty/Pk	Catalogue Number
Clear PET Membrane			
0.4 µm	6-well	48	PCHT06H48
1 µm			PTRP06H48
3 µm			PCSP06H48
0.4 µm	12-well	48	PCHT12H48
1 µm			PTRP12H48
3 µm			PCSP12H48
0.4 µm	24-well	48	PCHT24H48
1 µm			PTRP24H48
3 µm			PCSP24H48

Translucent PET Membrane

0.4 µm	6-well	48	PTHT06H48
1 µm			PLRP06H48
3 µm			PTSP06H48
5 µm			PTMP06H48
0.4 µm	12-well	48	PTHT12H48
1 µm			PLRP12H48
3 µm			PTSP12H48
5 µm			PTMP12H48
0.4 µm	24-well	48	PTHT24H48
1 µm			PLRP24H48
3 µm			PTSP24H48
5 µm			PTMP24H48

Cleaning Products

Tergazyme® enzyme detergent 1.8 kg Z742918

Alconox® enzyme detergent 1.8 kg Z724914

Water sterile-filtered, BioReagent, suitable for cell culture 500 mL W3500-500mL

Search 'Milli-Q® water' at [SigmaAldrich.com](https://www.sigmaaldrich.com) to find a wide variety of water and water purification products.

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