INSTRUCTION MANUAL

EVM-EL-03-XX-XX

STX HTS EVOM™ Electrode for TEER
in Multiwell Plates

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CONTENTS

ABOUT THIS MANUAL

The following symbols are used in this guide:

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

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

NOTES and TIPS contain helpful information.

Fig. 1—The handheld STX HTS electrode is keyed to fit into specific multiwell plates. This one is designed for use with the Millipore 96-well plate.

INTRODUCTION

The STX HTS EVOM™ electrode is a miniature, compact electrode designed for measurement of transepithelial electrical resistance (TEER) in high throughput screening (HTS) cell culture plates in combination with WPI's EVOM™ Manual. The STX HTS electrode exhibits nearly the same reproducibility for tissue resistance measurements (< ±5Ω) as WPI's Endohm chambers.

It also has the advantage of being able to perform resistance measurements directly in an HTS plate, base plate common or divided, reducing the possibility of contamination as well as mechanical damage to the cultured cells. The STX HTS employs a specially shaped adapter that fits precisely to the specific manufacturer's plate type. This ensures the consistent electrode positioning every time the electrode

is inserted into a well, significantly improving reproducibility over hand-held or manually placed electrodes.

Another important feature is the miniaturization of the electrode tips, only around 1.8 mm in diameter, yet stronger and more durable by design.

Although the STX HTS electrodes were developed to meet the demands of the pharmaceutical industry's HTS protocols, it is also a useful tool for the academic research scientist who requires precision, reproducibility, decreased exposure to contamination and minimal disturbance to cells. An example of such an application is the long-term study of TEER changes due to chemical or other factors.

Choose your STX HTS electrode based on the multiwell plate you are using.

Parts List

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

(1) **STX HTS** electrode, one of the following:

- **EVM-EL-03-02-05** STX HTS EVOM™ Electrode for MatTek 96
- **EVM-EL-03-03-02** STX HTS EVOM™ Electrode for Millipore 96
- **EVM-EL-03-03-03** STX HTS EVOM™ Electrode for Falcon 24
- **EVM-EL-03-03-04** STX HTS EVOM™ Electrode for Corning 24
- **EVM-EL-03-03-05** STX HTS EVOM™ Electrode for Corning 96

(1) Instruction Manual is available online at https://www.wpiinc.com/manuals.

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 11 of this manual. Please contact WPI Customer Service if any parts are missing at 941.371.1003 or customerservice@wpiinc.com.

Returns: Do not return any goods to WPI without obtaining prior approval (RMA # required) and instructions from WPI's Returns Department. Goods returned (unauthorized) by collect freight may be refused. If a return shipment is necessary, use the original container, if possible. If the original container is not available, use a suitable substitute that is rigid and of adequate size. Wrap the instrument in paper or plastic surrounded with at least 100mm (four inches) of shock absorbing material. For further details, please read the section entitled "Claims and Returns" on page 11 of this manual.

INSTRUMENT DESCRIPTION

WPI offers STX HTS electrodes for five different multiwell plate types, which are each keyed to fit the wells in the specific plates.

Fig. 2—This STX HTS is designed for use with a Millipore 96 multiwell plate.

Fig. 3—Left to right, these are for the Falcon 24, Corning 24, MatTek 96, and Corning 96.

The STX HTS electrodes have a connector that plugs into the EVOM™ Manual and the EVOM3. For use with older EVOM™ meters, use the STX4 Electrode Adapter Cable (WPI #: EVM-AC-02-01-01), which is not included.

Fig. 4—STX HTS connector.

Positioning the Electrode

The electrode fits like a lock and key inside the well plate and maintains a steady and consistent position inside the well plate. The wider base goes in the center (for example, apical side or top of the membrane). After placing the electrode correctly or in the right orientation, the electrode should be able to stand on its own on top of the sample well while taking readings.

OPERATIONS

Refer to the instruction manual of EVOM™ Manual meter for complete instructions on taking measurements.

Taking Resistance Measurements

1. Disconnect the EVOM™ Manual from the charger and turn the power on. Select the Resistance or Ohms mode on the EVOM™ Manual.
2. Disinfect the electrode.
3. Connect the electrode to the meter.
4. Precondition the electrode in media for 10 minutes.
5. Perform the measurements. To obtain the actual tissue reading, subtract the blank resistance value.
6. Clean the electrode, and let it air dry. Then, store the electrode.

Taking Voltage Measurements

1. Sterilize the electrode.
2. Connect the electrode to the EVOM™ Manual and leave the power off.
3. Equilibrate the electrode in growth media while connected to the EVOM™ Manual.

   NOTE: The electrode tips must be kept immersed in solution (media) to equilibrate for at least 3–4 hours in order to achieve a stable voltage.
4. Disconnect the EVOM™ Manual from the charger.
5. Turn the Power on.
6. Select the PD (potential difference) mode on the EVOM™ Manual.
7. Remain in the PD mode for 15 minutes while the electrode remains immersed in the media.
8. Measure the voltage in the media and zero the millivolt offset.
9. Measure the voltage/PD of an empty insert or membrane with media and without any cells. To obtain the actual tissue reading, subtract the blank insert voltage value from sample voltage values.
10. Clean the electrode, and let it air dry. Then, store the electrode.

MAINTENANCE

Electrodes must be properly cleaned, disinfected and stored.

Cleaning/Maintaining the STX HTS Electrodes

1. After you finish taking measurements for the day, soak (immerse) the electrode tips in 70% ethanol or isopropanol for 5–10 minutes.
2. Rinse the electrode tips with distilled water and allow them to air dry.
3. Store the electrodes dry and in a place away from light (or with minimal light).
4. When the electrodes are used frequently, soak the electrode tips once a week in a 1% Tergazyme solution for 15 minutes. Then, rinse them well with distilled water. Enzol may be used as an alternative to Tergazyme. This step may be done just before disinfection and before beginning an experiment.
5. Chloride the electrode tips by keeping the electrode tips immersed in 3–6% sodium hypochlorite (bleach), followed by rinsing with DI water. Perform this chloriding step once every 1–2 weeks when the electrode is used frequently or when the electrode is being used after long-term storage (more than 1 week).

Disinfecting the STX HTS Electrode

The STX HTS electrode is resistant to most methods of low temperature (or room temperature) chemical disinfection. The electrode tips may be disinfected by immersing the tips in a disinfection solution. Wipe the rest of the electrode with a paper towel that has been sprayed with a cleaning solution. Ortho-phthaladehyde (Cidex OPA or Rapicide OPA), 70% ethanol or 70% isopropyl alcohol can be used. A solution of 5% sodium hypochlorite (undiluted household bleach) is also a good choice. After disinfecting, rinse the electrode tips in sterile water, buffer or media before using the electrode to take sample measurements.

CAUTION: NEVER leave the electrode in alcohol (isopropanol or ethanol) for more than 30 minutes at a time. Continuously soaking the electrode in alcohol can shorten the electrode's life. Electrode tips that show signs of peeling or tearing may need to be replaced.

CAUTION: Do NOT use ammonia for cleaning.

CAUTION: Do NOT immerse the electrode head or any cable or connector portion in liquid. Only the electrode tip portion may be immersed in a liquid.

CAUTION: Do NOT sand or abrade the electrode tip surfaces.

Sterilizing the STX HTS Electrodes

The STX HTS electrodes are non-sterile as supplied. Acceptable low temperature sterilization methods for the electrodes include gamma irradiation and ethylene oxide gas (ETO). Chemical disinfection is generally considered adequate before using the STX HTS electrode for experiments.

CAUTION: Do NOT apply any heat or flame to any portion of the electrode or the electrode blades.

Storing the Electrodes

After a daily use, wash and store the electrodes dry and away from light. Regular cleaning and maintenance are critical for proper functionality and for maintaining the functional life of the electrode.

CAUTION: Always lift the electrode by the body, never by the cable.

CAUTION: Limit the liquid immersion or liquid spray level somewhere below the maximum level indicated by the arrows. You don't want the liquid to get inside and reach up to the cables or connectors. You can wipe the rest of the electrode with a paper towel sprayed with isopropanol or ethanol. (Do not spray onto the electrode directly.)

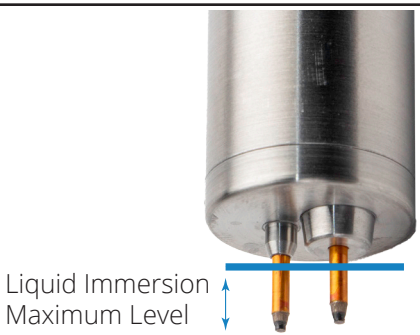

Fig. 5—Don't allow any liquid to pass the line indicated in this image.

APPENDIX A: COMPATIBLE PLATES

The STX HTS is compatible with the following plates.

STX HTS for MatTek 96 (EVM-EL-03-02-05)

Part #	Description	Pore Size	Membrane
CCI96-PET-0.4	PermaCell Culture Insert Plate PET, 96-well	0.4µm	PET
CCI96-PET-CL-0.4	PermaCell Culture Insert Plate, 96-well, Clear	0.4µm	PET

STX HTS for Millipore (EVM-EL-03-03-02)

Part #	Description	Pore size	Membrane
PSHT004R1	96-well plate	0.4um	PCF
PSRP004R1	96-well plate	1.0um	PET
PSHT004R5	96-well plate	0.4um	PCF
PSRP004R5	96-well plate	1.0um	PET
PSHT004S5	96-well plate	0.4um	PCF

STX HTS for Falcon (EVM-EL-03-03-03)

Part #	Description	Pore Size	Membrane
351181	Corning Falcon HTS Multiwell Insert System	1.0µm	PET
351183	Corning Falcon HTS Multiwell Insert System	3.0µm	PET
351185	Corning Falcon HTS Multiwell Insert System	8.0µm	PET
354803	Corning BioCoat HTS Fibrillar Collagen Multiwell Insert System	1.0µm	PET
354804	Corning BioCoat HTS Fibrillar Collagen Multiwell Insert System	1.0µm	PC

STX HTS for Corning 24 (EVM-EL-03-03-04)

Part #	Description	Pore Size	Membrane
3378	HTS Transwell-24	0.4µm	PET
3379	HTS Transwell-24	0.4µm	PET
3396	HTS Transwell-24	0.4µm	
3397	HTS Transwell-24	0.4µm	
3398	HTS Transwell-24	3.0µm	
3399	HTS Transwell-24	3.0µm	

STX HTS for Corning 96 (EVM-EL-03-03-05)

Part #	Description	Pore Size	Membrane
3380	HTS Transwell-96 System	1.0µm	PET
3392	HTS Transwell-96 System	1.0µm	PET
7369	HTS Transwell-96 System	0.4µm	PET
3381	HTS Transwell-96 System	0.4µm	PC
3391	HTS Transwell-96 System	0.4µm	PC
3385	HTS Transwell-96 Well Plate	3.0µm	PC
3386	HTS Transwell-96 Well Plate	3.0µm	PC
3387	HTS Transwell-96 Well Plate	5.0µm	PC
3388	HTS Transwell-96 Well Plate	5.0µm	PC
3374	HTS Transwell-96 Well Plate	8.0µm	PET
3384	HTS Transwell-96 Well Plate	8.0µm	PET

FREQUENTLY ASKED QUESTIONS (FAQ)

Is the electrode compatible with EVOM™ Manual, EVOM3 and EVOM2 meters?

The electrode can be connected directly with EVOM3 and EVOM™ Manual meters. The electrode is expected to provide the best user experience with an EVOM™ Manual or EVOM3 meter, because these next generation meters have the ability to store the data of a 96-well plate in Microsoft® Excel format.

The electrode can also be used with older version EVOM2 meters. To connect the electrode with an EVOM2, you need the STX4 Electrode Adapter Cable (WPI #: EVM-AC-02-01-01), which is not included.

What are the recommended volumes to be used with the electrode for TEER measurement?

WPI recommends using a minimum of 150 µL apical (top of the membrane) and 400 µL basolateral (bottom of the membrane) volumes with the 96-well insert plates.

How do I test if the electrode is working properly?

The electrode can be tested for proper functionality by measuring the resistance values of potassium chloride (KCl) dissolved in deionized water. Prepare 40, 80, 160 mM KCl solutions.

Without any membranes, below are the expected values when measured in a 24-well plate format. Make sure adequate volume is added to the well of the well plate so that the electrode tip remains immersed in the liquid during the test (See Fig. 5). Drastically different values outside this range can be indicative of erratic behavior.

Molarity	40 mM	80 mM	160 mM
Acceptable Range	140 ± 40 Ω	75 ± 23 Ω	38 ± 12 Ω

NOTE: The values can fluctuate in 40 mM KCl solution. The values should be stable in 80 and 160 mM KCl solutions. With increasing KCl concentrations, the resistance should show a decrease.

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 90 days* 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 typically warranted for 30 days only from the date on which the customer receives these items. EVOM™ electrodes, except STX4 (EVM-EL-03-03-01, are warranted for 90 days.*

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