

STX3

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2. Rinse the electrode in a sterile electrolyte solution similar to the experimental cell culture medium or in 0.1 M KCl or 0.15 M NaCl solution.
3. For *resistance* measurements, the electrode is now ready to use.
4. For *voltage* measurements, allow the electrode to equilibrate in the sterile electrolyte for 15 minutes. Then, adjust the "voltage zero" on the **EVOM** to balance any residual offset between the two probes.

The electrode can be left in the UV hood to keep it sterile.

NOTE: when the electrode is exposed to strong visible or UV light, a dark colored oxide film will slowly form on the electrode surface. This film normally will not affect the performance of the electrode. To avoid the formation of the film, shield the electrode from strong light.

STORAGE OF THE ELECTRODE

Short term storage (use within a week or so): Immerse the electrode tip in electrolyte solution. Make sure the electrode cable plug is connected to the electrode port on the **EVOM** meter so that the system is internally short-circuited and electrode symmetry is maintained.

Long term storage (greater than a week or two): When storing for long periods of time, the electrode should be rinsed with distilled water and stored dry and in the dark.

WARRANTY

WPI (World Precision Instruments, Inc.) warrants to the original purchaser that this equipment, including its components and parts, shall be free from defects in material and workmanship for a period of 30 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.

STX3

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.



STX3 Electrode

for use with EVOM Epithelial Voltohmmeter

INSTRUCTION MANUAL

Serial No. _____

070601



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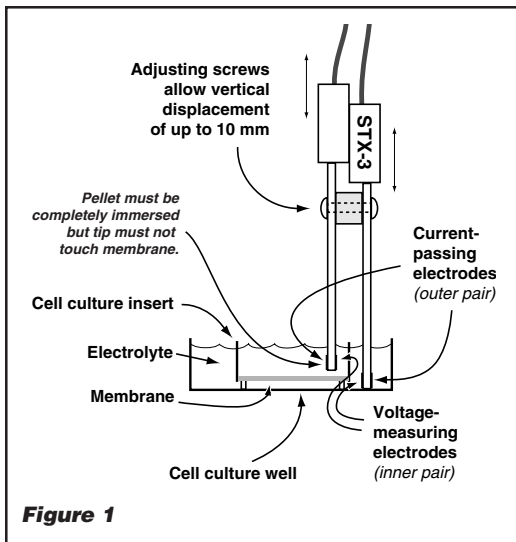
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The **STX3** electrode is designed to facilitate measurements of membrane voltage and resistance (TEER) of cultured epithelia in tissue culture wells. The electrode incorporates an adjustable pair of probes, 4 mm wide and 1 mm in thickness. This adjustable design allows the vertical position of the probes to

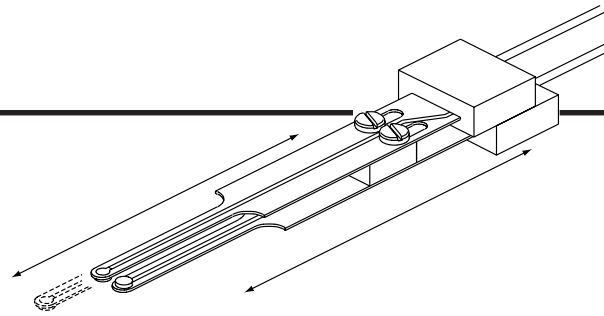
be optimized for any type of cell culture insert and distinguishes the **STX3** from the **STX2** electrode. The two probes may also be completely separated, if desired. Each probe has an outer and an inner electrode. The outside electrodes are small silver pads that pass current through the membrane sample. They are referred to as current electrodes and are connected to the I_1 and I_2 pins of the telephone-type plug connector. The internal electrodes are small Ag/AgCl pellet voltage sensors. They are referred to as voltage electrodes and are connected to the V_1 and V_2 pins of the telephone-type plug connector which plug into the **EVOM** or **EVOMX**.

The adjustable feature of the probes ensures proper positioning between the electrodes and the cell layer in the cup during the trans membrane measurement. The electrode should be set-up so that one probe, designated as the external electrode, is longer and touches the bottom of the dish containing the external culture media while the shorter (internal electrode) is adjusted so that it is prevented from reaching the membrane or bottom of the tissue culture insert. In addition, by positioning the longer tip so that it touches the bottom of the dish each time, the reproducibility of positioning the electrode and therefore the reading on successive measurements, is significantly improved. Even small differences in the apparent fluid resistance may occur if the depth to which the electrode tips are immersed varies.

ELECTRODE SET-UP

- Before using the electrode, the distance between the two electrodes should be adjusted so that when the longer electrode touches the bottom of the cell culture well, the shorter electrode should neither touch the membrane of the cell culture insert by its tip, nor let the silver/silver-chloride pellet be exposed to the air (see Fig. 1). To adjust the distance of the electrode, loosen the screws on the electrode probe as shown in Fig. 2, adjust to the desired distance and tighten before use. For most brands of cell culture inserts, a difference of 2.5 mm between the two electrodes will work best. However, some, such as the Transwell insert, may need a slightly longer distance.
- For *resistance* measurement, the electrode can be used directly out of dry storage without any preconditioning.

Figure 2



- For *voltage* measurements, the electrodes need to be equilibrated to eliminate any offset before use. Electrochemists have long used the following equilibration technique to assure voltage stability and a low inter-electrode potential difference:

The **STX3** voltage electrode pairs are shorted together internally when they are connected to the instrument and the instrument power switch is off. The user may therefore immerse the electrode in electrolyte solution (e.g., 0.1 – 0.15 M KCl) with the electrodes connected to the **EVOM** (Power Off) to allow the probes to equilibrate. With the voltage electrode connector pins thus short-circuited for several hours, the asymmetrical potential difference across the two voltage electrodes is reduced. The inter-electrode DC potential will be a few millivolts or less and quite stable.

The following table lists the recommended equilibration time before using the electrode.

Table 1: STX-3 Electrode Equilibration Time

| Electrode Condition: | Equilibration Time: |
|--------------------------------|---------------------|
| Never tested for voltage drift | 24 hours |
| Stored dry | 24 hours |
| Stored in electrolyte solution | 2 hours |

INSTRUCTIONS FOR USE

- Sterilize the electrodes, if desired. (See the Sterilization under Maintenance for detailed instructions)
- Place the electrode into the well so the tips just touch the bottom of the wells *without flexing* the electrode (Fig. 1). Ensure that the longer electrode lightly touches the bottom of the outer well and that it remains vertical. *To obtain reproducible results in the same cup, the position of the electrodes must remain constant. To improve the reproducibility and stability of the measurement, it is important to steady the electrode while measuring.*
- When moving the electrodes from one sample cup to another, it is best not to rinse the electrodes with distilled water. Silver/silver chloride electrodes may take several minutes to recover from exposure to distilled water, during which time the potential may drift by a few millivolts. 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.

MAINTENANCE

Cleaning

With use, the electrode surface can become coated with foreign materials. This build-up, or contamination, will degrade the performance of the system resulting in slow or erratic voltage readings.

We recommend three steps to clean the electrode:

- Using a cotton swab soaked with alcohol, lightly rub the surface of the electrode.
- If there is no improvement after following step 1 above, then soak the electrode in undiluted household bleach solution for 3 minutes. Rinse with water immediately after soaking. *Ensure that only the electrode tip is exposed to bleach.* Do not permit the bleach to touch the upper part of the electrode. If additional cleaning is required, go to Step 3.
- Lightly* rub the voltage electrode (silver pellet on the inner surface near the electrode tips) of the **STX3** with the 600-grade Ultra Fine sandpaper provided. See Fig. 3. Only a very thin surface layer of the pellet should be removed. Repeated rubbing will eventually remove the Ag/AgCl pellets. When rubbing no longer improves the voltage readings, the electrode should be replaced. In the absence of 600-grade sandpaper, an ink eraser may be substituted to clean the electrodes.

CAUTION: Only the plastic part of the tip can be sanded. Do not sand any metal part of the tip or damage to the electrode will result. Do not sand the current electrode.

Sterilizing the STX3 Electrode

The **STX3** electrodes are non-sterile as supplied. The electrode cannot be sterilized by autoclaving. They may be sterilized using alcohol (one of the most common methods), ethylene oxide, UV, or a bactericide, e.g., Cidex Plus (WPI #7364) or Sporicidin.

CAUTION: Do not flame electrodes. Doing so will cause them to melt and invalidate any warranty.

Typical Sterilization Protocol Using Alcohol

Do not leave the electrode in alcohol for more than 30 minutes each time. Continuously soaking the electrode in alcohol will weaken the protective coating on the electrode and shorten its lifetime.

In a laminar flow hood:

- Immerse the electrodes in 70% ethanol for 15 minutes. Allow them to air dry for 15 seconds.

