

Transepithelial Electrical Resistance (TER) Tight Junction Testing

- ** The probe must be immersed in solution before reading a measurement
- ** The probe should not touch the cells on the membrane
- ** All samples and solutions should be at room temp before testing

Calibrate the Millicell-ERS

1. Place the probe in a 100ml beaker with 20ml of 70% EtOH for 8-10min for sterilization
2. While sterilizing, take samples, PBS, and any media out to acclimate to room temp
3. Take probe out of the EtOH and air dry for 10-30sec and place in the sterile PBS
4. Turn the mode switch to “V” and turn the power on
5. Allow the reading to drift for 5min or until there is a stable reading. Adjust the system with a screwdriver until the meter shows a voltage reading of 0.0
6. Turn the power off and switch the mode to “R”. Turn the probe on and measure the resistance of the PBS (usually 1-4 ohms)

Measuring TER

1. Place the probe in the first “blank” sample without touching the insert membrane. Press the “Measure” button (top left) and allow the reading to stabilize before recording the TER
 - Keep the probe perpendicular to the sample plate
 - With the 12-well plates, the longer probe can touch the bottom of the well without touching the insert membrane
 - Keep the probe off of the well walls and not having fluid climb the probe for a more stable and accurate reading
2. Remove from the well and move to the next blank. Run all blanks before testing any wells with cells. Next test samples with cells but no treatment, and then treated cells to minimize contamination from the probe.
3. After the TER readings are recorded place the probe into the EtOH and clean the area. Remove the probe from the EtOH and place in the PBS for storage.

Changing Media (During monolayer growth)

1. Always remove the lower well media before removing the insert well media. This will prevent any cellular transfer from the top well to the bottom well.
2. Using a pipettman and P-1000 tip, extract the bottom well media. The insert well will rise and slide out of the way enough to place the tip at the bottom of the well.

3. Using the same tip, remove the top well media. Place the tip on the top well's side wall near the membrane but not touching the membrane, and extract as much media as possible. Keeping the tip on the side of the well gives you more control of the pipette tip and will remove more media by working with hydrostatic forces.
4. Refill both wells with fresh media

Viewing the membrane after TER study

1. Remove all of the media from both the top and bottom wells
2. Add a few drops of 4% paraformaldehyde to the top well.
3. Remove the paraformaldehyde from the top well after 30 sec of inundation
4. Remove the top well from the plate and using a razor blade cut the membrane from the bottom of the well following the wall of the well
5. Place the membrane face up on a glass slide for staining or viewing under a microscope.