ADHERENT CELL SAMPLES

Adherent cells can be grown directly on a flat E-chip using typical tissue culture conditions. For the following steps, place each E-chip into a well (of a flat bottom culture dish) that has been pre-filled with fluid. Then, transfer the E-chip between wells during each solution change or rinse step as described below.

- 1. Glow discharge or plasma clean the E-chips prior to use.
- 2. Soak the E-chip in a solution of 0.01% poly-L-lysine for 5 minutes, then rinse 3 times in water.
- 3. Place each E-chip into a well containing cell growth media with the E-chip oriented such that the flat surface is facing up.
- 4. Add a droplet of cell suspension to each well containing an E-chip .
- 5. Place the well plate in the incubator and allow cells to attach for several minutes
- 6. Check periodically until 4-5 cells are observed sticking to the SiN window.
- 7. Wait 5-10 minutes, then transfer each E-chip to a new well containing media (300 μL).
- 8. Allow the cells to adhere and grow under normal incubation conditions.
- 9. Transfer the E-chips between wells to rinse, label, fix, or stain the cells as desired.
- 10. Load the flow E-chip into the Poseidon holder (Live cells can be loaded in the Poseidon holder for immediate imaging, or fixed with glutaralehyde, for

imaging at a later time). Plasma clean the spacerE-chip and load it into the holder. Add a dropletof cell-compatible buffer, and then place the flatE-chip containing the cells on top. Secure the tip, andoperate with continuous flow to maintain hydration.

GOLD FIDUCIALS

Fiducial markers can be introduced onto the E-chip surface by applying gold nanoparticles to the surface of the E-chip prior to sample loading.

- 1. Glow discharge the spacer E-chips (2 minutes in either a glow discharge or plasma cleaner is fine)
- Place the spacer E-chips back onto the gel-pack to hold them in place (make sure the E-chips are membrane-side up, do not allow the membranes to contact the sticky gel surface).
- Dispense 0.5-1 microliter of the aqueous gold solution onto the surface of the E-chips and let the droplets sit for 5 minutes. (Note, do not let the droplets dry out on the E-chip).
- 4. Remove the excess droplet from the E-chip (either by wicking it off with filter paper or removing it with the pipet).
- 5. Dispense 0.5-1 microliter of the water onto the surface of the E-chip and let sit for 5 minutes.
- 6. Pick up the E-chip from the gel-pack and hold it by the edges with the carbon fiber tweezers.
- Use compressed air to blow the excess liquid off the E-chip, and dry the surface.
- 8. Plasma clean or glow discharge the E-chip a second time to stick the gold nanoparticles to the surface.