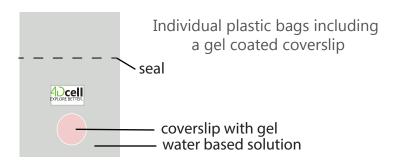
# Microscope round coverslips coated with polyacrylamide gels

#### Materials included



## Extra materials recomended and not included

Tweezers
PBS
Distilled water
Micropipette 200 µL
Micropipette 1000 µL
35 mm Petri dish or multi-well plate

Note: Always employ aseptic techniques to maintain sterility of coverslips and other materials.

### A. UNPACKAGING



Cut along the seal line to open the plastic bag.

(Be careful that the coverslip with the gel is not along the seal line)



Remove the coverslip from the bag using tweezers and carefully place it in the center of a Petri dish or multi-well plate. (be aware that the gel coated side of the coverslip is facing the 4Dcell logo sticker)



Safely discard the water based solution and plastic bag.

### **B. SEEDING CELLS**



Rinse the gel coated coverslip twice with PBS.



In the Petri dish or multi-well plate add 2 mL of pre-warmed cell culture medium and incubate for 15 minutes at  $(37^{\circ}C, 5\%)$  CO<sub>2</sub>, 95% humidity.



Resuspend 200,000 to 300,000 cells in 200  $\mu$ L of cell culture medium per coverslip<sup>(1)</sup>.



Pipet drop by drop 200  $\mu$ L of cell suspension on top of each patterned coverslip (throughout the whole gel surface) and incubate for at least 1 hour (depending on cell type) at (37°C, 5%) CO<sub>2</sub>, 95% humidity<sup>(2)</sup>.



Wash unattached cells with pre-warmed culture medium or sterile PBS.



Add 2 mL of pre-warmed culture medium per slide and incubate cells overnight for future analysis (3).

- (1) Please note that the number of cells depends on the confluence you want to attain, you should optimize the process according to your experiments and cell type used.
- (2) When adding the cell suspension, try to create a domelike bubble on top of the coverslip so the cell suspension is placed only on top of the gel. If needed, before adding the cell suspension, carefully transfer the gel-containing coverlip to an empty Petri dish or well and then pipet the cell suspension on top. This guarantees that the cell suspension will not spread out of the coverslip.
- (3) Plating of cells is optimized for HeLa cell line. Therefore, the number of cells added, the adhesion, and/or culture times can vary. Make sure the gels do not dry at any moment during the protocol.

