

READ THIS BEFORE USE

The following instructions are essential for successful handling.

Necessary Equipment Supplied by the User

Pipet Tips and Pipets

The μ -Slide Chemotaxis is **ONLY** compatible with the following pipet tips and products with identical geometry:

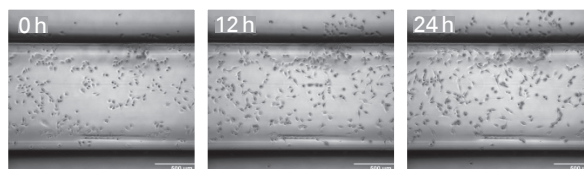
- Greiner Bio-One 739261, 739280, 739290, 772288
- Axygen T-200-C, TR-222-C, TR-222-Y
- Starlab TipOne RPT S1161-1800
- Sorenson BioScience MultiFit Tip 10470, 10590, 15270T, 30450

Using other tips will skew your results!

- Use a pipet that is serviced and calibrated routinely.

Video Microscopy

- **Ensure that video microscopy equipment is available.** Live cell imaging is absolutely necessary for documenting the cell migration for assay analysis.
- During video microscopy, ensure that your cells are incubated at the optimal temperature, humidity, and gas concentration.



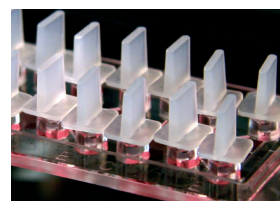
Before Starting

Read and Follow the Protocol

- Read through the entire instructions provided in Application Note 17. Follow the steps precisely and **do not change any of them.**

Practice General Handling

- To practice handling and pipetting, **use the practice kit** that comes with the free samples of the μ -Slide Chemotaxis.
- Avoid air bubbles inside the pipet tip.
- Make sure to fully fill the pipet adapters on the slide to avoid trapped air bubbles.



Important Handling Information for the μ -Slide Chemotaxis

Read **BEFORE** using the ibidi μ -Slide Chemotaxis

Perform Overnight Gas Equilibration

- **Gas equilibration of the labware and media being used is absolutely necessary before starting the assay.** Any air bubbles in your slide will ruin the experiment.
- For gas equilibration, place all the required media, the μ -Slide Chemotaxis, and the caps/plugs in an incubator the day before use (under the incubation conditions of the assay—typically 37°C, 5% CO₂). **Incubate overnight.**
- You may leave the μ -Slide Chemotaxis and the caps/plugs enclosed in the sterile package during gas equilibration.
- Slightly loosen the screw top of the medium container to ensure gas exchange.

During the Experiment

Let the Slide Dry Completely After Coating

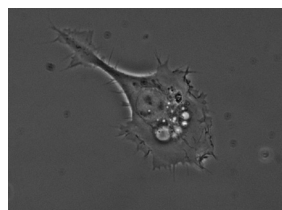
- If coating the slides yourself, the chambers of the μ -Slide Chemotaxis must be allowed to dry completely before cell seeding. Remove as much water/buffer after the coating as possible and let the μ -Slide Chemotaxis dry in the sterile bench.
- Make sure to perform gas equilibration of the μ -Slide Chemotaxis again after coating.

Maintain High Humidity to Avoid Evaporation

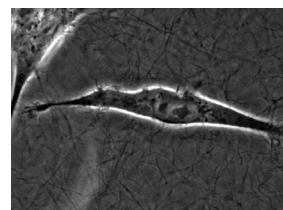
- To avoid evaporation during cell attachment, **always maintain high humidity in your sample by keeping the μ -Slide Chemotaxis constantly in a wet chamber** after cell seeding. The humidity of the incubator alone is not sufficient!
- Do not frequently open your incubator during the process of cell attachment.

3D Chemotaxis Assays

For 3D cell culture, cells should be surrounded by a gel matrix. Adherent cells might sink down and attach to the 2D surface. In your analysis, please make sure to distinguish clearly between the cells adhered to the 2D surface and the cells actually in 3D.



Example of a HT-1080 cell with a flat, expanded morphology when attached to the substrate in a 2D environment.



HT-1080 cell with a spindle-shaped morphology when cultured in a 3D matrix.



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