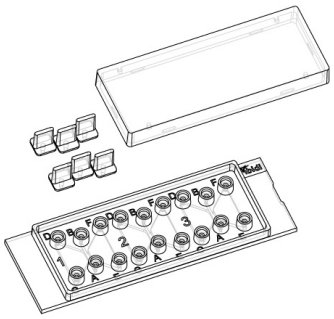


The ibidi product family is comprised of a variety of µ–Slides and µ–Dishes, which have all been designed for high–end microscopic analysis of fixed or living cells. The high optical quality of the material is similar to that of glass, so you can perform all kinds of fluorescence experiments with uncompromised resolution and choice of wavelength.

The µ–Slide Chemotaxis is a tool for investigation of chemotaxis and migration of non-adherent cells in 3D gel matrices and adherent cells on 2D surfaces. The chamber's geometry is optimized for analyzing chemotaxis by video microscopy. The linear concentration profile which is required for chemotactical movement is generated by diffusion and stable for at least 48 hours.

Please read the following Application Note for more detailed information: Application Note 17 "2D and 3D Chemotaxis Assays using µ–Slide Chemotaxis":

Material

ibidi µ–Slides, µ–Dishes, and µ–Plates are made of a plastic that has the highest optical quality. The polymer coverslip on the bottom exhibits extremely low birefringence and autofluorescence, similar to that of glass. Also, it is not possible to detach the bottom from the upper part. The µ–Slides, µ–Dishes, and µ–Plates are not autoclavable, since they are only temperature–stable up to 80°C/175°F. Please note that gas exchange between the medium and incubator's atmosphere occurs partially through the polymer coverslip, which should not be covered.

Optical Properties ibidi Polymer Coverslip	
Refractive index n_D (589 nm)	1.52
Abbe number	56
Thickness	No. 1.5 (180 µm)
Material	polymer coverslip

Please note! The ibidi polymer coverslip is compatible with certain types of immersion oil only. A list of suitable oils can be found on page 2.

Coating your µ–Slide Chemotaxis

Detailed information about coatings is provided in Application Note 08 "Cell culture coating".

Shipping and Storage

The µ–Slides, µ–Dishes and µ–Plates are sterilized and welded in a gas-permeable packaging. The shelf life under proper storage conditions (in a dry place, no direct sunlight) is listed in the following table.

Conditions	
Shipping conditions	Ambient
Storage conditions	RT (15-25°C)

Shelf Life of Different Surfaces	
ibiTreat, Glass Bottom, ESS	36 months
Collagen, Poly-L-Lysine	18 months

Geometry

Geometry of the µ–Slide Chemotaxis	
Chambers on slide	3
Volume per chamber	130 µl
Observation area	2×1 mm^2
Distance between chambers	18.5 mm
Total height with plugs	12 mm
Volume chemoattractant	30 µl
Bottom matches coverslip	No. 1.5

µ–Slide Surfaces

Depending on your cells and special application you will need µ–Slides with different surfaces. If you do not require any special adhesion molecules for your application, the best choice will be ibiTreat, a tissue culture treated surface.

The µ–Slide Chemotaxis is also provided with a Collagen coated surface (not a 3D gel). Only the observation area is coated to mediate cell adhesion for possible 2D experiments. Such an adhesion substrate has been shown to stimulate the adhesion and growth of various cell lines in µ–Slides. A high quality Collagen IV solution (Corning #356233) is used to pre-coat the slides.

Tip:

The ibiTreat surface is very hydrophilic which facilitates filling the structure with aqueous gels for 3D assays.

Seeding Cells

Detailed information on correct slide handling is provided in Application Note 17 "2D and 3D Chemotaxis Assays using µ–Slide Chemotaxis".

Here are the short steps for cell seeding and conducting chemotaxis experiments:

1. Prepare your cell suspension as usual. Use cell suspension of approx. 3 ×10^6 cells/ml (final in gel).
2. Bring cell suspension into a gel*. [1]
3. Close filling ports of the large reservoirs by plugs.
4. Apply 6 µl gel onto one filling port of the side channel. Do not inject the gel directly.
5. Aspirate 6 µl of air from the opposite filling port. The gel–cell mixture will be flushed into the channel.
6. Close the two filling ports of the channel.
7. Remove all plugs from the filling ports of the large reservoirs.
8. Incubate the slide inside a sterile and humid atmosphere to minimize evaporation until the gel is formed. Make sure evaporation is low by using a sterile 10 cm Petri dish with extra wet tissue around the slide.

Important!

The day before seeding place the cell medium and the µ–Slide into the incubator for equilibration. This will prevent the liquid inside the channel from emerging air bubbles over the incubation time.

Chemotaxis Experiment

1. After the gel matrix is solidified fill both reservoirs with 65 µl chemoattractant–free medium.
2. Fill 2 ×15 µl chemoattractant solution into one reservoir.
3. Close all filling ports with plugs.
4. Conduct video microscopy.
5. Track cells and analyze migration. Please visit www.ibidi.com for a software tool analyzing migrational data.

Cell seeding and conducting a chemotaxis experiment is described in detail in Application Note 17.

Perform control experiments (+;+) and (-;-) similarly.

Preparation for Cell Microscopy

To analyze your cells, no special preparations are necessary. Cells can be observed live, or fixed directly in the µ–Slide on an inverted microscope. You can use any fixative of your choice. The µ–Slide material is compatible with a variety of chemicals, e.g., acetone or methanol. Further specifications can be found at www.ibidi.com. Due to the thin bottom of only 180 µm, high resolution microscopy is possible.

Troubleshooting

Tips and tricks on handling and further troubleshooting is provided in Application Note 17 "2D and 3D Chemotaxis Assays Using µ–Slide Chemotaxis".

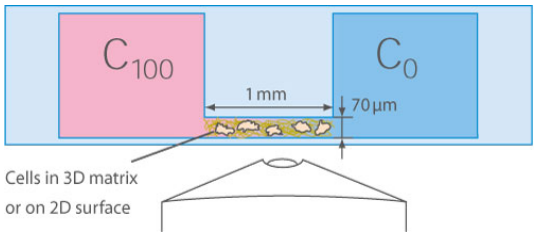
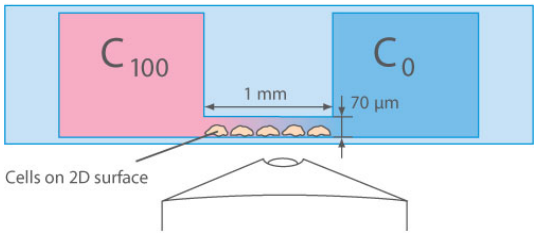
Immersion Oil

When using oil immersion objectives, use only the immersion oils specified in the table. The use of a non–recommended oil could lead to the damage of the plastic material and the objective.

Company	Product	Ordering Number
Zeiss	Immersol 518 F	(Zeiss) 444960
Zeiss	Immersol W 2010	(Zeiss) 444969
Leica	Immersion liquid	(Leica) 11513859

[1] Please find example protocols here:
Application Note 23 "3D Chemotaxis Protocol with Collagen I Gel for Dendritic Cells"
Application Note 26 "Fabrication of Collagen I Gels"

µ-Slide Chemotaxis Selection Guide

µ–Slide Chemotaxis	µ–Slide Chemotaxis 2D
Migration of non–adherent cells in 3D gel matrix.	Migration of strongly adherent cells on a flat surface.
Can also be used for a 2D chemotaxis experiment with adherent cells.	Only for 2D experiments.
For fast or slow migrating cells embedded in a 3D gel matrix (or adherent cells, respectively) e.g. neutrophils, lymphocytes and dendritic cells. A gel matrix is not part of the product.	For slow migrating, adherent cells, e.g. endothelial cells, cancer cells or fibroblasts.
Gradient is long term stable.	Gradient is long term stable.
Easy handling	Challenging handling
	

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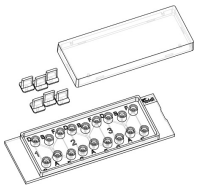
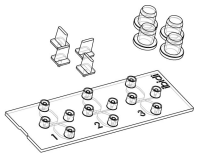
Ordering Information

µ–Slide Chemotaxis



Cat. No.	Description
80326	**µ–Slide Chemotaxis ibiTreat**: #1.5 polymer coverslip, tissue culture treated, sterilized
80322	**µ–Slide Chemotaxis Collagen IV***: #1.5 polymer coverslip, sterilized

µ–Slide Chemotaxis ^{2D}



Cat. No.	Description
80306	**µ–Slide Chemotaxis ^{2D} ibiTreat**: #1.5 polymer coverslip, tissue culture treated, sterilized
80302	**µ–Slide Chemotaxis ^{2D} Collagen IV***: #1.5 polymer coverslip, sterilized

* Surface coating of observation area. Does not contain a gel matrix.

For research use only!

Further technical specifications can be found at www.ibidi.com. For questions and suggestions please contact us by e-mail *info@ibidi.de* or by telephone +49 (0)89/520 4617 0. All products are developed and produced in Germany.