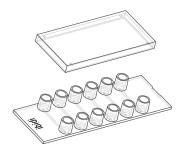
Instructions μ -Slide VI $^{0.1}$



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 VI $^{0.1}$ is designed for flow assays in a minimal volume. It can be connected to a pump and enables you to observe cells under flow conditions. The small dimensions offer you the possibility to work with a minimum of cells (e.g. mouse model). The convenient six channel format is ideal for the application of standard protocols for e.g. treatment, staining, and microscopy of living or fixed cells.

Material

ibidi μ –Slides, μ –Dishes, and μ –Plates are made of a plastic that has the highest optical quality. The bottom material 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 plastic bottom, which should not be covered.

Optical Properties ibidi Standard Bottom			
1.52			
56			
No. 1.5 (180 μm)			
microscopy plastic			

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

Geometry

The μ –Slide VI $^{0.1}$ provides a standard slide format according to ISO 8037/1. The lateral adapter to adapter distance of 9 mm (like 96 well plates) allows using multichannel pipettes.

Dimensions				
Number of channels	6			
Channel volume	1.7 µl			
Channel length	17 mm			
Channel width	1.0 mm			
Channel height	0.1 mm			
Adapters	Female Luer			
Volume per reservoir	60 μl			
Growth area	$0.17 \mathrm{cm}^2 \mathrm{per} \mathrm{channel}$			
Coating area using 1.7 µl	0.34 cm ² per channel			
Bottom matches coverslip	No. 1.5			

μ-Slide Surfaces

Depending on the type of cells and the special application you are using, 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.

We provide precoated μ –Slides with adhesion substrates like Collagen IV, Fibronectin, Poly–L–Lysin, and Poly–D–Lysin. Such adhesion substrates have been shown to stimulate the adhesion and growth of various cell lines in μ –Slides. Only high–quality substrates are used 1 .

The uncoated μ –Slide is manufactured from hydrophobic plastic. For the cultivation of most cell lines, it is indispensable to treat the uncoated μ –Slide with biopolymers, which mediate cell adhesion and growth.

¹Collagen IV, BD Cat.-Nr. 35 6233, Fibronectin, BD Cat.-Nr. 354008, Poly-L-Lysin, Sigma Cat.-Nr. P4832, Poly-D-Lysin, BD Cat.-Nr. 354210



Instructions µ-Slide VI 0.1

Coating your µ-Slide VI 0.1

The uncoated μ –Slide must be coated to promote cell adhesion. If you want to establish a certain coating to match your needs, we recommend testing your coating procedure on both uncoated and ibiTreat μ –Slides, since we have observed that some biomolecules adhere differently to hydrophobic and hydrophilic plastic surfaces.

- Prepare your coating solution according to the manufacturer's specifications or reference.
- Apply 1.7 μl per channel and leave at room temperature for at least 30 minutes.
- Aspirate the solution and wash with ultra-pure water. You can add the water into one channel end and simultaneously aspirate it on the other side. Let dry at room temperature.

Further information about coatings are provided in Application Note 08 Cell culture coating.

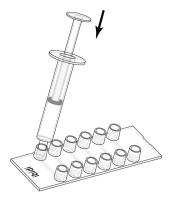
- Cover reservoirs with the supplied lid. Incubate at 37°C and 5 % CO₂ as usual.
- Await cell attachment in order not to flush out the cells. Afterwards fill each reservoir with 60 µl cell free medium.
- Connect the μ–Slide to the pump and conduct your perfusion experiment.

Tip:

The day before seeding the cells we recommend placing 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.

Filling and Handling

Filling the very small channels of μ –Slide VI $^{0.1}$, especially hydrophobic uncoated, by a normal pipette might be challenging. Please use a small volume syringe with a Luer tip for convenient filling the channels with coating solution or cell suspension.



Exchanging Medium

Aspirate both reservoirs and slowly fill 60 µl of fresh medium into each of the reservoirs. The equilibration of the liquid levels may take some minutes because of the small width of the channel.

Important!

Please note that the $\mu\text{--Slide VI}^{0.1}$ is not for use in static culture due to the small channel volume. Cultivation without perfusion is only possible by a medium exchange every few hours or by using a rocker which constantly generates a slight medium flow between the two reservoirs.

Seeding Cells

- Trypsinize and count cells as usual. Dilute the cell suspension to the desired concentration. Depending on your cell type, application of a $12-28 \times 10^5$ cells/ml suspension should result in a confluent layer within 2–3 days.
- Apply 1.7 μl cell suspension into the channel of the μ-Slide. Quick dispensing helps to avoid trapped air bubbles.

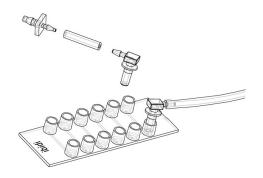
Flow Application

Detailed information about flow rates, shear stress, and shear rates is provided in Application Note 11 "Shear stress and shear rates" on www.ibidi.com.

Suitable flow kits (μ –Slide VI $^{0.1}$ + tubing and adapters) are also available (see page 3).

μ–Slide VI ^{0.1}

Instructions



Please contact us for recommended perfusion setups. ibidi provides a variety of pump systems.

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 μ m, high resolution microscopy is possible.

μ-Slide VI 0.1 Family

The μ -Slide VI $^{0.1}$ family is available with different surfaces. See table below for choosing your μ -Slide VI $^{0.1}$.



Ordering Number	Treatment or Coating	Characteristics
80666	ibiTreat, sterile	hydrophilic, tissue culture treated
80662	Collagen IV, sterile	protein coating
80663	Fibronectin, sterile*	protein coating
80664	Poly-L-Lysine, sterile	biopolymer coating
80661	uncoated, sterile	hydrophobic

^{*} available on request only

μ –Slide VI $^{0.1}$ Flow Kits

The μ -Slide VI $^{0.1}$ is available as flow kit with tubes and adapters.



Ordering Number	Treatment or Coating	Characteristics
80686	ibiTreat, sterile	hydrophilic, tissue culture treated
80682	Collagen IV, sterile	protein coating
80681	uncoated, sterile	hydrophobic



Instructions μ -Slide VI $^{0.1}$

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 46 17 0. All products are developed and produced in Germany. © ibidi GmbH, Am Klopferspitz 19, 82152 Martinsried, Germany.