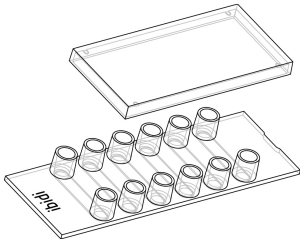


The ibidi product family is comprised of a variety of µ–Slides and µ–Dishes, which have all been designed for high–end microscopic analysis of fixed or living cells. The high optical quality of the material is similar to that of glass, so you can perform all kinds of fluorescence experiments with uncompromised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

The convenient six channel format of the µ–Slide VI 0.4 is ideal for static cell cultivation and the application of standard immunofluorescence protocols, like treatment, staining, and microscopy of living or fixed cells. Alternatively, the µ–Slide VI 0.4 can be connected to a pump and enables you to observe cells under flow conditions.

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	
Refractive index n_D (589 nm)	1.52
Abbe number	56
Thickness	No. 1.5 (180 µm)
Material	microscopy plastic

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

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.

Geometry of the µ–Slide VI 0.4

The µ–Slide VI 0.4 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	30 µl
Channel length	17 mm
Channel width	3.8 mm
Channel height	0.4 mm
Adapters	female Luer
Volume per reservoir	60 µl
Growth area	0.6 cm^2 per channel
Coating area using 30 µl	1.2 cm^2 per channel
Bottom matches coverslip	No. 1.5

Coating your µ–Slide VI 0.4

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 30 µl per channel and leave at room temperature for at least 30 minutes.

[1] Collagen IV: Corning #356233, Fibronectin: Corning #354008, Poly–L–Lysin: Sigma #P4832, Poly–D–Lysin: Corning #354210

- Aspirate the solution and wash with the recommended protein dilution buffer. You can add the buffer into one channel end and simultaneously aspirate it on the other side.
- Optionally, let dry at room temperature. Attention, some coating proteins might degenerate when drying!

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

Seeding Cells

- Trypsinize and count cells as usual. Dilute the cell suspension to the desired concentration. Depending on your cell type, application of a 3–7 × 10^5 cells/ml suspension should result in a confluent layer within 2–3 days.
- Apply 30 µl cell suspension into the channel of the µ–Slide. Quick dispensing helps to avoid trapped air bubbles.
- Cover reservoirs with the supplied lid. Incubate at 37°C and 5 % CO_2 as usual.
- Await cell attachment in order not to flush out the cells. Afterwards fill each reservoir with 60 µl cell free medium.

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.
Trapped air bubbles can be removed from the channel by inclining the µ–Slide and knocking at one edge.

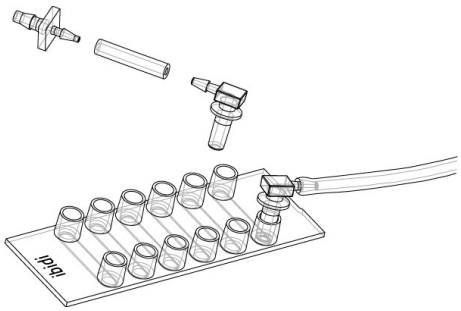
Exchanging Medium

Aspirate both reservoirs and fill slowly 120 µl of fresh medium into one of the reservoirs, which will replace the channel volume by gravity flow.

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 Tube Adapter Sets are also available (see page 3). They consist of a tubing (20 cm) with inner diameter of 1.6 mm and adapters for the connection between the ibidi µ–Slide (female Luer) and the tubing of the pump in use.



Please contact us for recommended perfusion setups. ibidi provides a variety of channel slides and 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 µm, high resolution microscopy is possible.

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
ibidi	Immersion Oil	(ibidi) 50101
Zeiss	Immersol 518 F	(Zeiss) 444960
Zeiss	Immersol W 2010	(Zeiss) 444969
Leica	Immersion liquid	(Leica) 11513859

µ–Slide VI 0.4 Family

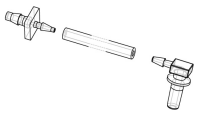
The µ–Slide VI 0.4 family is available with different surfaces. See table below for choosing your µ–Slide VI 0.4.

Ordering Number	Treatment or Coating	Characteristics
80606	ibiTreat, tissue culture treated, sterile	hydrophilic, tissue culture treated
80602	Collagen IV, sterile	protein coating
80603	Fibronectin, sterile*	protein coating
80604	Poly-L-Lysine, sterile	biopolymer coating
80605	Poly-D-Lysine, sterile*	biopolymer coating
80601	uncoated, sterile	hydrophobic

* available on request only

Tube Adapter Set

For the connection of the ibidi µ–Slides to any flow system suitable Tube Adapter Sets are also available. They consist of a tubing (20 cm) with inner diameter of 1.6 mm and adapters for the connection between the ibidi µ–Slide (female Luer) and the tubing of the pump in use.



Ordering Number	Product Name	Characteristics
10831	Tube Adapter Set	12 pcs, sterile

Selected References

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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.