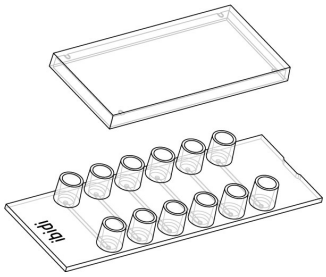


The ibidi product family comprises a variety of different shapes of µ–Slides and µ–Dishes which all have been designed for high–end microscopic analysis of fixed or living cells. The high optical quality of the material is similar to that of glass, so you can perform all kinds of fluorescence experiments 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 and µ–Dishes consist of a plastic with highest optical quality. The material exhibits extremely low birefringence and autofluorescence, both similar to that of glass. It is not possible to detach the bottom from the upper part. The µ–Slides and µ–Dishes are not autoclavable since they are temperature stable up to 80°C/175°F only. 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

Geometry

The µ–Slide VI 0.1 provides 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 cm^2 per channel
Coating area using 1.7 µl	0.34 cm^2 per channel
Bottom matches coverslip	No. 1.5

Immersion oil

When using oil immersion objectives, only the immersion oils specified in the table may be used. The use of different oil can lead to damages of the plastic material and the objective.

Company	Product	Ordering number
Cargille	type DF, Formula Code: 1261	(Cargille) 16242
Zeiss	518 F	(Zeiss) 444960
Olympus	50CC	(Olympus) 35506
Nikon	50 CCM DF	(Nikon) MXA 20351
Leica	immersion oil, low fluorescence	(Leica) 11513859

µ–Slide surfaces

Depending on your cells and special application you will need µ–Slides with different surfaces. If you do not need 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 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 cultivation of most cell lines it is indispensable to treat the uncoated µ–Slide with biopolymers which mediate cell adhesion and growth.

Coating your µ–Slide VI 0.1

The uncoated µ–Slide must be coated to promote cell adhesion. If you like to establish a certain coating for your demands we recommend testing your coating procedure on uncoated and ibiTreat µ–Slides, since we have observed

[1] Collagen 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

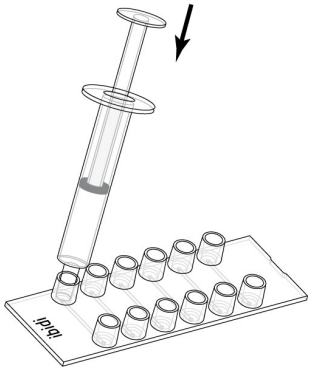
that some biomolecules adhere differently to hydrophobic or hydrophilic plastic surfaces.

- 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.

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.



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 × 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.
- 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.
- 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.

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. This may take some minutes because of the small width of the channel.

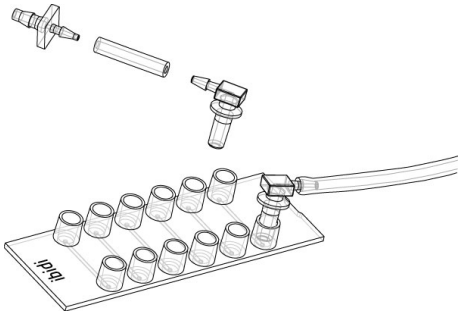
Important!

Please note that the µ–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.

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).



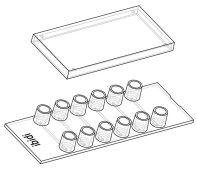
Please contact us for recommended perfusion setups. ibidi provides a variety of pump systems.

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 µm, 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.

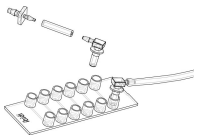


Ordering number	Treatment or Coating	Characteristics
80666	ibiTreat, sterile	hydrophilic, tissue culture treated
80662	Collagen IV, sterile	protein coating
80663	Fibronectin, sterile*	protein coating
80664	Poly-L-Lysine, sterile	biopolymer coating
80661	uncoated, sterile	hydrophobic

* 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

Selected cell tests on different surfaces

Many eukaryotic and bacterial cells have been tested by ibidi on the different surfaces of the µ-Slides. A variety of other cell lines like COS, CHO, HepG2, and NIH 3T3 were successfully grown by our customers.

	ibiTreat	Collagen IV	Fibronectin	Poly-L-Lysin	Poly-D-Lysin	uncoated
HUVEC	excellent	good	excellent	no cell growth	not done	no cell growth
Rat1	excellent	excellent	excellent	excellent	excellent	poor
HT1080	excellent	excellent	excellent	excellent	not done	poor
HeLa	excellent	excellent	excellent	excellent	not done	poor
Neuro2A	excellent	excellent	excellent	excellent	excellent	poor
PC12	good	excellent	excellent	excellent	excellent	no cell growth
Dictyostelium discoideum	not done	excellent	not done	not done	not done	excellent
Escherichia coli	excellent	not done	not done	excellent	not done	excellent

HUVEC = Human Umbilical Vein Endothelial Cells
Rat1 = Rat Fibroblast
HT1080 = Human Fibrosarcoma
HeLa = Human Cervix Adenocarcinoma
Neuro2A = Mouse Neuroblastoma
PC12 = Rat Pheochromocytom

Dictyostelium discoideum = strain wild type AX-2

Escherichia coli = strain MDG131

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.