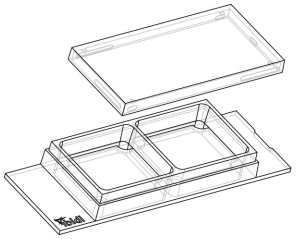


The ibidi product family is comprised of a variety of µ–Slides and µ–Dishes, which have all been designed for high–end microscopic analysis of fixed or living cells. The high optical quality of the material is similar to that of glass, so you can perform all kinds of fluorescence experiments with uncompromised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

The µ–Slide 2 well Ph+ (Phase contrast plus) is an array of 2 square fields where cells can be cultivated and investigated with microscopical methods. The µ–Slide 2 well Ph+ improves the optical quality of phase contrast microscopy. In contrast to the classic µ–Slide 2 well, the Ph+ version provides a special plate in the center of the wells. This plate suppresses the meniscus which is disturbing the phase contrast effect in normal open wells. Openings near the corners provide access to the wells for filling and aspirating liquids easily.

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

Geometry

The µ–Slide 2 well Ph+ provides a standard slide format according to ISO 8037/1.

Geometry of µ–Slide 2 well Ph+	
Number of wells	2
Dimensions of wells (w × l × h) in mm	21.2 × 23.3 × 3.0
Growth area per well	4.8 cm^2
Coating area per well	11.4 cm^2
Volume per well	1.5 ml
Liquid height	3.0 mm
Total height with lid	10.8 mm
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.

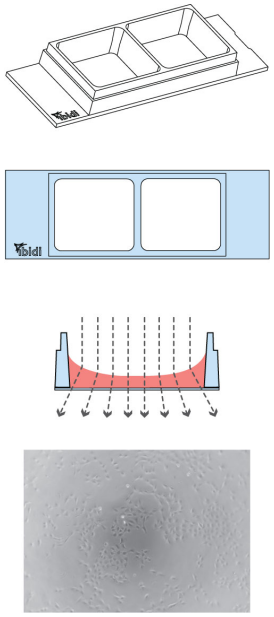
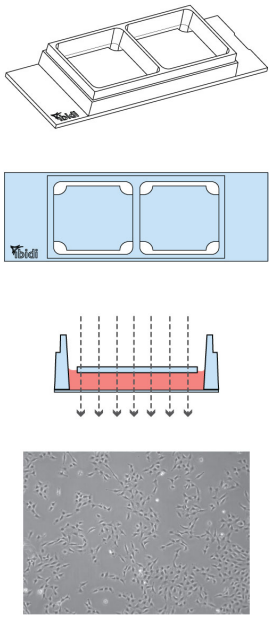
Coating your µ–Slide 2 well Ph+

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 1.5 ml per well and leave at room temperature for at least 30 minutes.
- Aspirate the solution and wash with ultra-pure water. Let dry at room temperature.

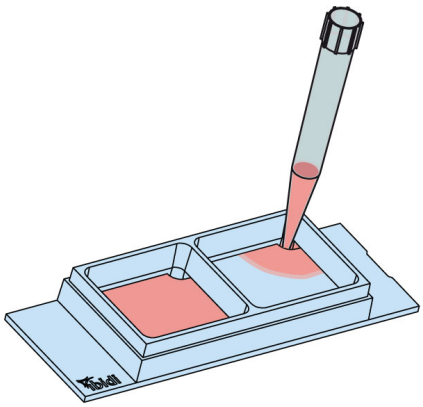
[1] Collagen IV, BD Cat.–Nr. 35 6233, Fibronectin, BD Cat.–Nr. 354008, Poly–L–Lysin, Sigma Cat.–Nr. P4832, Poly–D–Lysin, BD Cat.–Nr. 354210

µ–Slide 2 well Ph+ selection guide

µ–Slide 2 well	µ–Slide 2 well Ph+
Standard open wells for maximum sample access. Meniscus disturbs the beam path. Good phase contrast quality only in the center of each well	Special plate in the center of the wells suppresses meniscus formation. No meniscus – parallel beam path. For excellent phase contrast microscopy all over the wells.
	

Filling and Handling

Fill the wells by using a standard pipet. Inject the cell suspension directly into one of the openings. Medium exchange is easily done by aspirating the entire volume and refilling using 1.5 ml per well



Seeding Cells

- Trypsinize and count cells as usual. Dilute the cell suspension to meet the desired concentration. Depending on your cell type, the application of a 5-11 × 10^4 cells/ml suspension should result in a confluent layer within 2–3 days.
- Pipet 1.5 ml cell suspension into each well of the µ–Slide. Avoid shaking, as this will result in an inhomogeneous distribution of the cells.
- Cover the reservoirs with the supplied lid. Incubate at 37°C and 5 % CO_2 as usual.

Undemanding cells can be left in their seeding medium for up to three days and can grow to confluence there. However, the best results might be achieved when the medium is changed every 1–2 days. Carefully aspirate the old medium and replace it with 1.5 ml/well fresh 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 from emerging air bubbles over the incubation time.

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

µ–Slide 2 well Family

The µ–Slide 2 well family is available as open well and as a Ph+ version. See table below for choosing your µ–Slide 2 well.

µ–Slide 2 well

Ordering Number	Treatment or Coating	Characteristics
80286	ibiTreat, sterile	hydrophilic, tissue culture treated
80282	Collagen IV, sterile	protein coating
80283	Fibronectin, sterile*	protein coating
80284	Poly-L-Lysine, sterile	biopolymer coating
80285	Poly-D-Lysine, sterile*	biopolymer coating
80281	uncoated, sterile	hydrophobic

* available on request only

µ–Slide 2 well Ph+

Ordering Number	Treatment or Coating	Characteristics
80296	ibiTreat, sterile	hydrophilic, tissue culture treated
80292	Collagen IV, sterile	protein coating
80293	Fibronectin, sterile*	protein coating
80294	Poly-L-Lysine, sterile	biopolymer coating
80295	Poly-D-Lysine, sterile*	biopolymer coating
80291	uncoated, sterile	hydrophobic

* available on request only

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.