

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 4 well<sup>Ph+</sup> (Phase contrast plus) is an array of 4 square fields where cells can be cultivated and investigated with microscopical methods. The μ-Slide 4 well<sup>Ph+</sup> improves the optical quality of phase contrast microscopy. In contrast to the classic μ-Slide 4 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

## 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<sup>1</sup>.

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.

<sup>1</sup>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

## Geometry

The μ-Slide 4 well<sup>Ph+</sup> provides a standard slide format according to ISO 8037/1.

### Geometry of μ-Slide 4 well

Number of wells	4
Dimensions of wells (w × l × h) in mm	21.2 × 11.0 × 3.0
Growth area per well	2.2 cm <sup>2</sup>
Coating area per well	5.9 cm <sup>2</sup>
Volume per well	700 μl
Liquid height	3.0 mm
Total height with lid	10.8 mm
Bottom matches coverslip	No. 1.5

## Coating your μ-Slide 4 well<sup>Ph+</sup>

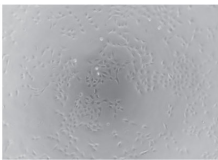
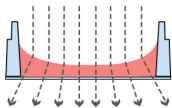
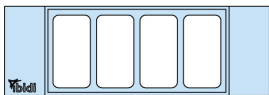
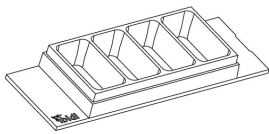
The uncoated μ-Slide must be coated to promote cell adhesion. If you want to establish a certain coating to match your needs, we recommend testing your coating procedure on both uncoated and ibiTreat μ-Slides, since we have observed that some biomolecules adhere differently to hydrophobic and hydrophilic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

- Prepare your coating solution according to the manufacturer's specifications or reference.
- Apply 700 μl 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.

## μ-Slide 4 well Ph+ selection guide

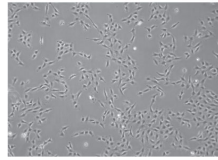
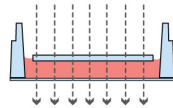
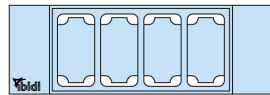
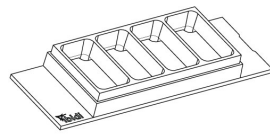
### μ-Slide 4 well

Standard open wells for maximum sample access. Meniscus disturbs the beam path. Good phase contrast quality only in the center of each well



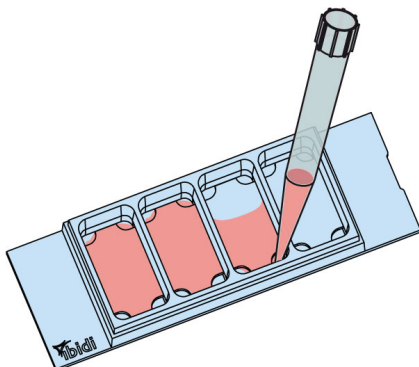
### μ-Slide 4 well Ph+

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 700 μl 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  $5-11 \times 10^4$  cells/ml suspension should result in a confluent layer within 2–3 days.
- Pipet 700 μl 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<sub>2</sub> 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 700 μl/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](http://www.ibidi.com). Due to the thin bottom of only 180 μm, high resolution microscopy is possible.

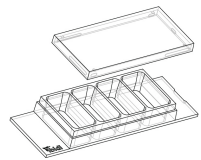
## Immersion Oil

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 4 well Family**

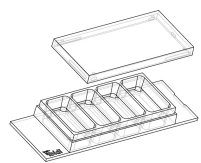
The μ-Slide 4 well family is available as open well and as a Ph+ version. See table below for choosing your μ-Slide 4 well.



Ordering Number	Treatment or Coating	Characteristics
80426	ibiTreat, sterile	hydrophilic, tissue culture treated
80422	Collagen IV, sterile	protein coating
80423	Fibronectin, sterile*	protein coating
80424	Poly-L-Lysine, sterile	biopolymer coating
80425	Poly-D-Lysine, sterile*	biopolymer coating
80421	uncoated, sterile	hydrophobic

\* available on request only

**μ-Slide 4 well<sup>Ph+</sup>**



Ordering Number	Treatment or Coating	Characteristics
80446	ibiTreat, sterile	hydrophilic, tissue culture treated
80442	Collagen IV, sterile	protein coating
80443	Fibronectin, sterile*	protein coating
80444	Poly-L-Lysine, sterile	biopolymer coating
80445	Poly-D-Lysine, sterile*	biopolymer coating
80441	uncoated, sterile	hydrophobic

\* available on request only

**For research use only!**

Further technical specifications can be found at [www.ibidi.com](http://www.ibidi.com). For questions and suggestions please contact us by e-mail [info@ibidi.de](mailto:info@ibidi.de) or by telephone +49 (0)89/520 46 17 0. All products are developed and produced in Germany.  
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