Instructions





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 -Plate 96 Well allows you to perform high resolution microscopy in a standard multi-well format. For less well-to-well crosstalk in fluorescence microscopy this imaging plate is made out of a black plastic material.

Material

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

Optical Properties ibidi Polymer Coverslip		
Refractive index n _D (589 nm)	1.52	
Abbe number	56	
Thickness	No. 1.5 (180 μm)	
Material	polymer coverslip	

Please note! The ibidi polymer coverslip is compatible with certain types of immersion oil only. A list of suitable oils can be found on page 3.

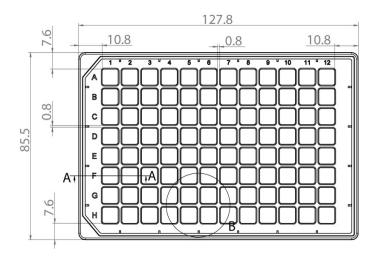
Shipping and Storage

The μ -Slides, μ -Dishes and μ -Plates are sterilized and welded in a gas-permeable packaging. The shelf life under proper storage conditions (in a dry place, no direct sunlight) is listed in the following table.

Conditions		
Shipping conditions	Ambient	
Storage conditions	RT (15-25°C)	
Shelf Life of Different Surfaces		
ibiTreat, Glass Bottom, ESS	36 months	
Collagen, Poly-L-Lysine	18 months	

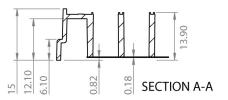
Geometry

The μ -Plate 96 Well provides standard geometry and numbering (A-H, 1-12). The bottom of the μ -Plate 96 Well provides a high accuracy.



The μ -Plate 96 Well meets all important values of the ANSI/SBS Standards (1-2004, 2-2004, 3-2004 and 4-2004).

Dimensions of the µ–Plate 96 Well in mm		
Length	127.8	± 0.2
Width	85.5	± 0.2
Height with lid	17.2	± 0.4
Height without lid	15.0	± 0.4
Well to well distance	9.0	± 0.1
Well clearance	0.82	± 0.1

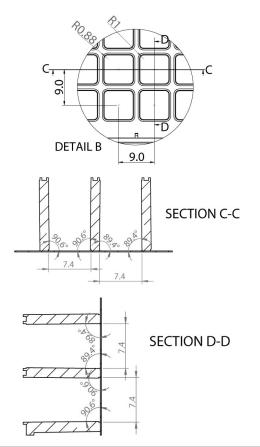




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Single well parameters of the μ -Plate 96 Well		
Single well dimensions	$7.4 \times 7.4 \pm 0.15 \text{ mm}$	
Single well depth	$12.9 \pm 0.2 \text{ mm}$	
Volume	300 µl	
Growth area	0.56 cm ²	
Coating area using 300 µl	2.35 cm^2	
Accuracy of the μ–Plate 96 Well bottom		
Inner well flatness	$\pm 5 \mu m$	
Whole plate flatness	$\pm 25 \ \mu m$	
Bottom matches coverslip	No. 1.5	

For manufacturing process reasons, the well wall angles differ slightly. The details are shown in the graphics below.



Surface

The μ -Plate 96 Well is available with ibiTreat and uncoated surface. The ibiTreat surface is a physical treatment and optimized for adhesion of most cell types. Many cell lines as well as primary cells were tested.

A specific coating of the μ -Plate 96 Well can be done yourself following the procedure in section Coating your μ -Plate 96 Well.

Coating your µ-Plate 96 Well

The uncoated μ -Plate 96 Well 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 -Plate 96 Well, since we have observed that some biomolecules adhere differently to hydrophobic or hydrophilic plastic surfaces.

- Prepare your coating solution according to the manufacturer's specifications or reference.
- Apply $300 \,\mu$ l in each well. The coating area using $300 \,\mu$ l is $2.35 \,cm^2$.
- Follow your coating protocol.

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

Remove the Protection Film before Usage

The bottom of the μ -Plate 96 Well is covered with a film to protect the optical quality of the plastic surface. Please pull off the protection film before usage!

Seeding Cells

- Trypsinize and count cells as usual. Dilute the cell suspension to the desired concentration. Depending on your cell type, application of a $2-5 \times 10^4$ cells/ml suspension should result in a confluent layer within 2–3 days.
- Apply 300 µl cell suspension into each single well. Avoid shaking as this will result in inhomogeneous distribution of the cells.
- Cover the μ -Plate 96 Well with the supplied lid. Incubate at 37°C and 5 % CO₂ as usual.

Tip:

We recommend not to fill more than $600 \,\mu$ l into the μ -Plate 96 Well in order to avoid liquid contacting the lid.

Undemanding cells can be left in their seeding medium for several days and grow to confluence there. However, best



results might be achieved when the medium is changed every 2–3 days. Carefully aspirate the old medium and replace it by 300 µl fresh medium.

Tip:

You can stack the μ -Plates to save space in you incubator. This will not affect cell growth. We recommend making batches with up to 6 plates, due to stability reasons.

Preparation for Cell Microscopy

For analyzing your cells no special preparations are necessary. Cells can be observed live or fixed directly in the μ -Plate preferably on an inverted microscope. You can use any fixative of your choice. The μ -Plate 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 nonrecommended oil could damage 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



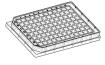
Ordering Information

The ibidi μ -Plates are available with the following surfaces.

µ–Plate 24 Well

	Cat. No.	Description
100000	82406 82401	μ–Plate 24 Well ibiTreat : #1.5 polymer coverslip, tissue culture treated, sterilized μ–Plate 24 Well Uncoated : #1.5 polymer coverslip, hydrophobic, sterilized

µ-Plate 96 Well



Cat. No.	Description
89626	μ–Plate 96 Well ibiTreat : #1.5 polymer coverslip, tissue culture treated, sterilized
89621	μ –Plate 96 Well Uncoated: #1.5 polymer coverslip, hydrophobic, sterilized

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. © ibidi GmbH, Am Klopferspitz 19, 82152 Martinsried, Germany.