

Instructions μ -Dish 50mm, low



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 –Dish $^{50\text{mm}, low}$ allows you to perform high resolution microscopy in a 50 mm Petri–dish with 9 mm walls. The low height makes high numerical apertures of Köhler illumination possible and provides large access for micromanipulation. The lid can be closed to hinder evaporation during long term experiments.

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 polymer coverslip, 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/	
	polymer coverslip	

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.

Geometry

Geometry of the µ–Dish 50mm, low		
Diameter dish	50 mm	
Volume	3 ml	
Growth area	7.0 cm^2	
Diameter growth area	30 mm	
Coating area using 700 µl	7.9 cm^2	
Height with / without lid	12 mm / 9 mm	
Bottom matches coverslip	No. 1.5	

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 Storage conditions	Ambient RT (15-25°C)	
Shelf Life of Different Surfaces		
ibiTreat, Glass Bottom, ESS Collagen, Poly-Lysine Fibronectin	36 months 18 months 4 months	

Surface and Coating

The μ –Dish 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 for good cell growth. Uncoated is a very hydrophobic surface and allows no direct cell growth. It is suitable for specific coatings or suspension cells.

The uncoated $\mu\text{-Dish}$ must be coated to promote cell adhesion. If you like to establish a particular coating for your demands we recommend to test your coating procedure on uncoated and ibiTreat $\mu\text{-Dishes},$ 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. Prepare your μ–Dish, ibiTreat or uncoated. Adjust the concentration to a coating area of 7.9 cm² and 700 μl.
- Apply 700 µl into the growth area. Make sure that the entire bottom is covered with liquid easily tilting

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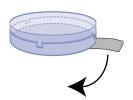
or shaking the μ -Dish. Put on the lid and leave at room temperature for at least 30 minutes.

 Aspirate the solution and wash. Optionally, let dry at room temperature.

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

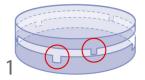
Protection Film

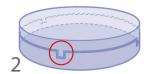
Remove the protection film before usage!



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

Using The Lid





- 1. open position, easy opening
- 2. close position, for long term studies, minimal evaporation

Seeding cells

Depending on your cell type, application of a $4-9 \times 10^4$ cells/ml suspension should result in a confluent layer within 2–3 days.

- Trypsinize and count cells as usual. Dilute the cell suspension to the desired concentration.
- Apply 700 μl cell suspension into the inner well of the μ-Dish. Avoid shaking as this will result in inhomogeneous distribution of the cells. After cell attachment add additionally 2.3 ml of pure medium to ensure optimal grow conditions.
- Cover the μ –Dish with the supplied lid. Incubate at 37°C and 5 % CO₂ as usual.

We recommend not to fill more than 3 ml into the μ -Dish in order to avoid the 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 up to 3 ml fresh medium.

Tip:

You can stack the μ –Dishes to save space in your incubator. This will not affect cell growth. We recommend making batches with up to 6 μ –Dishes, due to stability reasons. Placing the μ –Dishes into larger Petri dishes simplifies transport and prevents evaporation, heat loss, and contamination when the incubator is opened.

Minimizing Evaporation

Using the μ –Dish with a closed lid, the evaporation in an incubator system with 37°C and 95% humidity is around 1% per day. Using the μ –Dish with a closed lid in a 37°C heating system with low humidity (between 20% and 40%), the evaporation is around 10% per day. For reducing the evaporation down to 1% per day in all systems, we recommend sealing the lid with ibidi Anti–Evaporation Oil (50051).

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

Preparation for Cell Microscopy

To analyze your cells no special preparations are necessary. Cells can be observed live or fixed directly in the $\mu\text{-Dish}$ preferably on an inverted microscope. You can use any fixative of your choice. The $\mu\text{-Dish}$ material is compatible



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with a variety of chemicals, e.g. Acetone or Methanol. Further specifications can be found at www.ibidi.com. Due to the thin bottom high resolution microscopy is possible.

For optimal results in fluorescence microscopy and stor-

age of stained probes, ibidi provides a mounting medium optimized for $\mu\text{-Dishes}$ and $\mu\text{-Slides}$ (ibidi Mounting Medium, 50001).



Instructions

$\mu ext{-Dish}^{\,50\text{mm, low}}$

μ-Dish 50mm, low Family

The $\mu\text{-Dish}^{50\text{mm, low}}$ is available with the ibiTreat surface and uncoated.

μ-Dish ^{50mm, low}



Cat. No.	Description	Characteristics
81136	μ – Dish ^{50mm, low} ibiTreat : \emptyset 50 mm, low wall (3 ml volume), #1.5 polymer coverslip, tissue culture treated	hydrophilic, sterilized
81131	μ –Dish ^{50mm, hlow} Uncoated: \emptyset 50 mm, low wall (3 ml volume), #1.5 polymer coverslip	hydrophobic, sterilized

μ-Dish ^{35mm, low}



Cat. No.	Description	Characteristics
80136	μ–Dish ^{35mm, low} ibiTreat : Ø 35 mm, low wall (800 μl volume), #1.5 polymer coverslip, tissue culture treated	hydrophilic, sterilized
80131	μ–Dish ^{35mm, low} Uncoated : Ø 35 mm, low wall (800 μl volume), #1.5 polymer coverslip	hydrophobic, sterilized

μ–Dish ^{35mm, high}



Cat. No.	Description	Characteristics
81156	μ – Dish ^{35mm, high} ibiTreat : \emptyset 35 mm, high wall (2 ml volume), #1.5 polymer coverslip, tissue culture treated	hydrophilic, sterilized
81151	μ –Dish ^{35mm, high} Uncoated: \emptyset 35 mm, high wall (2 ml volume), #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.