# µ–Dish <sup>50mm, low</sup>



# Instructions



The ibidi product family is comprised of a variety of  $\mu$ -Slides and  $\mu$ -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  $\mu$ -Dish<sup>50mm, low</sup> 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  $\mu$ -Slides,  $\mu$ -Dishes, and  $\mu$ -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  $\mu$ -Slides,  $\mu$ -Dishes, and  $\mu$ -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 <sub>D</sub> (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.

#### Geometry

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

#### Shipping and Storage

The  $\mu$ -Slides,  $\mu$ -Dishes and  $\mu$ -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			

## **Surface and Coating**

The  $\mu$ -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  $\mu$ -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  $\mu$ -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 cm<sup>2</sup> and 700 μl.
- Apply 700 µl into the growth area. Make sure that the entire bottom is covered with liquid easily tilting

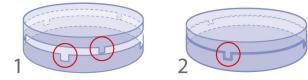


or shaking the  $\mu$ -Dish. Put on the lid and leave at room temperature for at least 30 minutes.

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

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

## 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  $\mu$ -Dish with the supplied lid. Incubate at 37°C and 5 % CO<sub>2</sub> as usual.

# We recommend not to fill more than 3 ml into the $\mu$ -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  $\mu$ -Dishes to save space in your incubator. This will not affect cell growth. We recommend making batches with up to 6  $\mu$ -Dishes, due to stability reasons. Placing the  $\mu$ -Dishes into larger Petri dishes simplifies transport and prevents evaporation, heat loss, and contamination when the incubator is opened.

## Preparation for Cell Microscopy

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

For optimal results in fluorescence microscopy and storage of stained probes, ibidi provides a mounting medium optimized for  $\mu$ -Dishes and  $\mu$ -Slides (ibidi Mounting Medium, 50001).

## **Minimizing Evaporation**

Using the  $\mu$ -Dish with a closed lid, the evaporation in an incubator system with 37°C and 95% humidity is around 1% per day. Using the  $\mu$ -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).



# Instructions

#### **Immersion Oil**

When using oil immersion objectives with the ibidi Polymer Coverslip, use only the immersion oils specified in the table below. The use of any non–recommended oil could damage the ibidi Polymer Coverslip. The resulting leakage may harm objectives and microscope components. All immersion oils that are not listed in the table below should be considered as non–compatible.

Company	Product	Ordering No.	Lot Number	Test Date
ibidi	ibidi Immersion Oil	50101	16-12-27	01/2017
Zeiss	Immersol 518 F	444960	160706	01/2017
Zeiss	Immersol W 2010	444969	101122	04/2012
Leica	Immersion Liquid	11513859	n.a.	03/2011
Cargille	Type A	16482	100592	01/2017
Cargille	Type HF	16245	92192	01/2017
Olympus	Silicone Immersion Oil	SIL300CS-30CC	N4190800	01/2017
Carl Roth	Immersion oil	X899.1	414220338	01/2017



# Instructions

#### µ–Dish<sup>50mm, low</sup> Family

The  $\mu$ -Dish<sup>50mm, low</sup> is available with the ibiTreat surface and uncoated.

## µ–Dish <sup>50mm, low</sup>

Cat. No.	Description	Characteristics
81136	<b>μ–Dish</b> <sup>50mm, low</sup> <b>ibiTreat</b> : Ø 50 mm, low wall (3 ml volume), #1.5 polymer coverslip, tissue culture treated	hydrophilic, sterilized
81131	<b>μ–Dish</b> <sup>50mm, hlow</sup> <b>Uncoated</b> : Ø 50 mm, low wall (3 ml volume), #1.5 polymer coverslip	hydrophobic, sterilized

## µ–Dish <sup>35mm, low</sup>

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

# µ–Dish <sup>35mm, high</sup>

Cat. No.	Description	Characteristics
81156	$\mu$ -Dish <sup>35mm, high</sup> ibiTreat: $\emptyset$ 35 mm, high wall (2 ml volume), #1.5 polymer coverslip, tissue culture treated	hydrophilic, sterilized
81151	<b>µ–Dish</b> <sup>35mm, high</sup> <b>Uncoated</b> : $\emptyset$ 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.