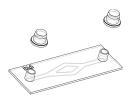


#### Instructions

#### μ-Slide y-shaped



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  $\mu$ -Slide y-shaped can be easily connected to a pump via its Luer connectors. It enables you to grow cells under flow conditions with a bifurcation of 30 and 45 degree depending on the flow

direction you choose. It is meant as a simulation system for blood vessels where the reaction of cells to a stimulus of your choice can be observed in real-time.

#### Material

ibidi  $\mu$ -Slides,  $\mu$ -Dishes, and  $\mu$ -Plates are made of a polymer 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 intended for one-time use and are not autoclavable, since they are only temperature-stable up to  $80^{\circ}$ C/175°F. Please note that gas exchange between the medium and the 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.

## **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 Storage conditions	Ambient RT (15–25°C)	
Shelf Life		
ibiTreat	36 months	

#### Geometry

The  $\mu$ -Slide y-shaped provides a standard slide format according to ISO 8037/1.

Geometry of the	μ-Slide y-shaped
Outer dimensions	25.5 mm x 75.5 mm
Adapters	Female Luer
Volume per reservoir	60 µl
Channel volume	110 µl
Channel height	0.4 mm
Channel width	3 mm
Branching angles	$30^{\circ}$ and $45^{\circ}$
Growth area	$2.8 \text{ cm}^2$
Coating area using 110 µl	$5.6 \text{ cm}^2$
Bottom	No. 1.5 ibidi Polymer Coverslip

#### Surface

The tissue culture-treated ibiTreat surface is a physical surface modification and optimized for adhesion of most cell types. ibiTreat is our most recommended surface modification, because most adherent cells grow well on this hydrophilic version of the ibidi Polymer Coverslip, without the need for any additional coating.

#### Coating

Detailed information about coatings is provided in Application Note 08: Coating protocols for ibidi labware products.

In short, specific coatings are possible following this protocol:

1. Prepare your coating solution according to the manufacturer's specifications or reference.

## μ-Slide y-shaped

#### Instructions

- 2. Apply 110 µl and leave at room temperature for at least 30 minutes.
- 3. Aspirate the solution and wash with the recommended protein dilution buffer.
- 4. The μ-Slide y-shaped is ready to be used. Optionally let dry at room temperature. Attention, some coating proteins might degenerate when drying!

#### Tip:

Apply the coating solution using a 1000 µl pipette and by quick pipetting. Adding more liquid than needed (e.g. 200 µl) is filling the channel easier. Aspirate the surplus 90 µl from the Luer reservoirs after filling. For washing you can add the washing buffer into one reservoir and simultaneously aspirate it on the other side.

## **Seeding Cells**

- Trypsinize and count cells as usual. Dilute the cell suspension to the desired concentration. Depending on your cell type, application of a  $3-7 \times 10^5$  cells/ml suspension should result in a confluent layer within 2–3 days.
- Apply 200 μl cell suspension into the channel of the μ-Slide using a 1000 μl pipette.
- Remove the cell suspension from the reservoirs.
- Cover reservoirs with the supplied caps. Incubate at 37°C and 5 % CO<sub>2</sub> as usual.
- After cell attachment fill each reservoir with 60 μl medium for longer cultivation.
- The μ-Slide is now ready for applying flow conditions on the adherent cells. Don't trap air bubbles when plugging in the connecting tubes.

Depending on the cells we recommend to exchange the medium every day in static culture: Aspirate both reservoirs. Flush fresh medium inside the channel by slowly filling one reservoir with 400  $\mu$ l medium and removing the content of the reservoir from the other well, ensuring the channel is never dry. Leave both reservoirs filled with approx. 60  $\mu$ l each.

#### Tip:

The day before seeding the cells we recommend placing the cell medium and the  $\mu$ -Slide into the cell culture incubator for equilibration. This will prevent the liquid inside the channel from emerging air bubbles over the incubation time.

Quick dispensing of cell suspension helps to avoid trapped air bubbles and leads to maximal homogeneity of cell distribution.

#### **Microscopy**

To analyze your cells, no special preparations are necessary. Cells can be directly observed live or fixed, preferably on an inverted microscope. The bottom cannot be removed. For optimal results in fluorescence microscopy and storage of fixed and stained samples, ibidi provides a mounting medium (50001) optimized for  $\mu$ -Dishes,  $\mu$ -Slides, and  $\mu$ -Plates.

#### **Chemical Compatibility**

The following table provides some basic information on the chemical and solvent compatibility of the  $\mu$ -Slide y-shaped. For a full list of compatible solvents and more information on chemical compatibility, please visit the FAQ section on ibidi.com.

Chemical / Solvent	Compatibility
Methanol	yes
Ethanol	yes
Formaldehyde	yes
Acetone	yes, without lid
Mineral oil	no
Silicone oil	yes
Immersion oil	See <b>Immersion Oil</b> on page 3.

## **Connecting Tubing for Perfusion**

The  $\mu\text{-Slide}$  is fully compatible with the ibidi Pump System and other pump setups.

Detailed information about flow rates, shear stress, and shear rates is provided in Application Note 11 "Shear stress and shear rates" and and Application Note 18



#### Instructions

#### μ-Slide y-shaped

"Shear Stress and Shear Rates in  $\mu$ -Slide y-shaped". Suitable Tube Adapter Sets are also available (see page 4). They consist of a tubing (20 cm) with inner diameter of 1.6 mm and adapters for the connection between the ibidi  $\mu$ -Slide (female Luer) and the tubing of the pump in use.

- 1. Fill both Luer ports with cell-free medium until they are completely filled. This ensures air bubble-free connection of the tubing.
- 2. Prepare the perfusion system by 1) filling the tubing completely and 2) pinching off the tubing with a screw clamp or a hose clip.
- 3. Connect the male Luer ends of the clamped tubing to the Luer ports one at a time. Make sure not to trap air. Remove access culture medium with tissue.
- 4. Open the clamped tubing and conduct your perfusion experiment.

#### **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.

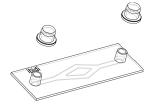
Company	Product	Ordering No.	Lot Number	Test Date
ibidi	ibidi Immersion Oil	50101	16-12-27	01/2017
Cargille	Type A	16482	100592	01/2017
Cargille	Type HF	16245	92192	01/2017
Carl Roth	Immersion oil	X899.1	414220338	01/2017
Leica	Immersion Liquid	11513859	n.a.	03/2011
Nikon	Immersion Oil F2 30cc	MXA22192	n.a.	01/2020
Nikon	Silicone Immersion Oil 30cc	MXA22179	20191101	01/2020
Olympus	Silicone Immersion Oil	SIL300CS-30CC	N4190800	01/2017
Zeiss	Immersol 518 F	444960	160706	01/2017
Zeiss	Immersol W 2010	444969	101122	04/2012



# μ-Slide y-shaped

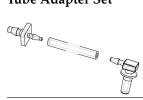
## Instructions

## **Ordering Information**



Cat. No.	Description
80126	$\mu$ -Slide y-shaped ibiTreat: #1.5 polymer coverslip, tissue culture treated, sterilized

#### **Tube Adapter Set**



Cat. No.	Description
10831	Tube Adapter Set: sterilized

## For research use only!

Further information 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.

© ibidi GmbH, Lochhamer Schlag 11, 82166 Gräfelfing, Germany.