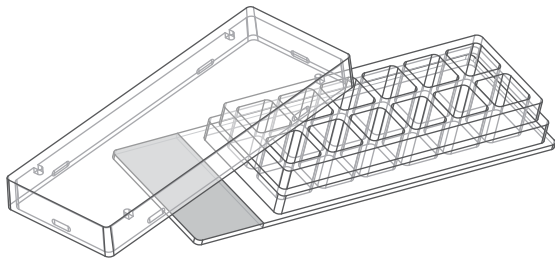


12 Well Chamber, removable

Instruction Manual



The 12 Well Chamber, removable is a silicone chamber mounted on a glass slide, designed for cell culture and immunofluorescence (IF) staining. The removable chamber allows standard cultivation, staining, and mounting procedures using coverslip sealing. After removing the chamber and mounting the glass slide with a coverslip, the sample can be analyzed using upright or inverted microscopes and is suitable for long-term storage. Compatible 24 × 60 mm coverslips are available from ibidi (#10811).

This document applies to the following product:

81201 **12 Well Chamber, removable**

Material

The 12 Well Chamber, removable consists of a self-adhesive silicone gasket mounted on a standard microscopy glass slide. The gasket is made of biocompatible silicone. Although both components are autoclavable and compatible with alcohols, reuse is not recommended. The glass slide features ground edges and a frosted labeling area.



CAUTION – Be cautious when handling ibidi labware products with a glass bottom! The glass coverslip or slide is fragile and can break easily. Handle these items carefully to prevent physical injury and damage to devices due to medium leakage.

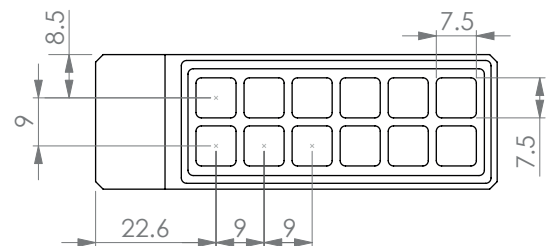
Shipping and Storage

This product is sterilized and sealed in a gas-permeable packaging. The shelf life under proper storage conditions (in a dry place, no direct sunlight) is outlined in the following table.

Conditions	
Shipping conditions	Ambient
Storage conditions	RT (15–25 °C)
Shelf Life	
Removable silicone chamber	36 months

Geometry

The 12 Well Chamber, removable provides standard slide format according to ISO 8037-1.



Specifications

Number of wells	12
Dimensions of wells (w × l × h)	7.5 × 7.5 × 8 mm
Volume per well	250 µl
Growth area per well	0.56 cm ²
Coating area per well	1.9 cm ²
Bottom (w × l × h)	Glass slide 26 × 76 × 1 mm
Height with lid	11 mm

Surface

The 12 Well Chamber, removable is a removable silicone chamber, mounted on an untreated standard microscopy glass slide. Washing steps (e.g., with PBS) before cell seeding helps remove glass dust, which enhances direct cell growth on the surface.

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Coating

Detailed information about coatings is provided in [Application Note 08: Coating Protocols for ibidi Labware](#).

In short, specific coatings are possible following this protocol:

1. Prepare your coating solution according to the manufacturer's specifications. Adjust the concentration to a coating area of 1.9 cm^2 and a volume of $250\text{ }\mu\text{l}$ per well.
2. Apply $250\text{ }\mu\text{l}$ coating solution per well. Make sure that the entire bottom is covered with liquid by slightly tilting or shaking the slide. Put on the lid and leave it at room temperature for at least 30 minutes.
3. Aspirate the solution and wash with the recommended protein dilution buffer.
4. The coated slide is ready to be used. Be aware that allowing the coated surface to dry out is not recommended, as some coating proteins may degrade upon drying.

Seeding Cells

1. Trypsinize and count the cells as usual. Dilute the cell suspension to the desired concentration. Depending on your cell type, application of a $4\text{--}9 \times 10^4$ cells/ml suspension should result in a confluent layer within 2–3 days.
2. Apply $250\text{ }\mu\text{l}$ cell suspension per well. Avoid shaking, as this will result in inhomogeneous cell distribution.
3. Cover the slide with the supplied lid. Incubate as usual (e.g., at 37°C and $5\% \text{ CO}_2$).

Insensitive cells can be left in their seeding medium for several days and grow to confluence there. However, optimal results might be achieved when the medium is changed every 2–3 days. For this, carefully aspirate the old

medium and replace it by $250\text{ }\mu\text{l}$ fresh medium per well.



NOTE – The 12 Well Chamber, removable is not recommended for high-resolution live cell imaging on inverted microscopes, as cells grow on a 1 mm thick microscopy glass slide.

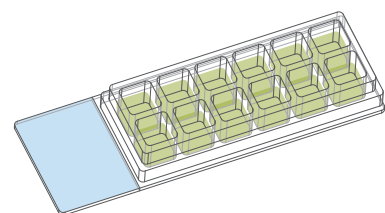
IF Handling and Processing

After cultivation, cells can be fixed and stained using one of the following two approaches. The slide is then mounted for microscopy.

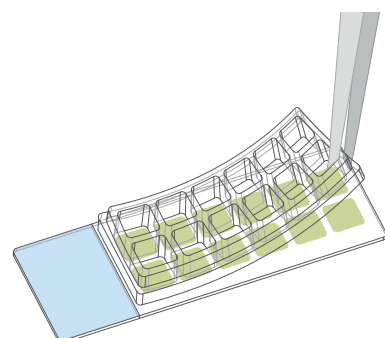
a) Single-Well Staining

All steps (fixation, permeabilization, staining, and washing) are performed within individual wells before removing the silicone gasket.

1. Perform your standard staining protocol directly in the individual wells.



2. Remove the silicone gasket by carefully lifting one edge with tweezers or by hand.



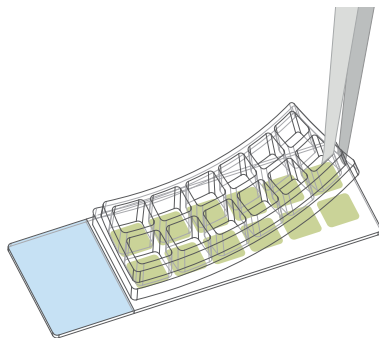
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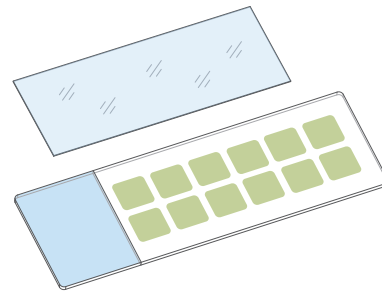
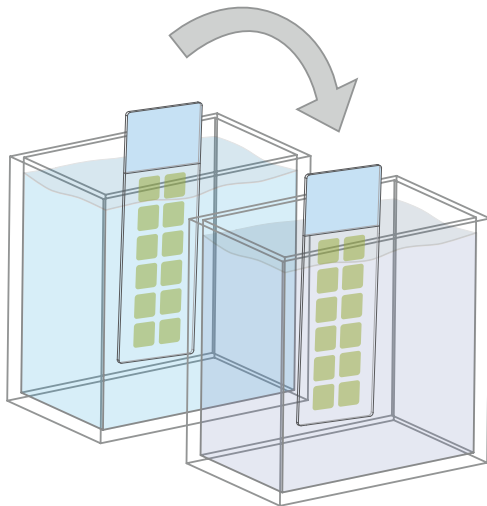
b) Parallel Staining

All steps (fixation, permeabilization, staining, and washing) are performed by dipping the entire slide in the respective solutions after removing the silicone gasket.

1. Remove the silicone gasket by carefully lifting one edge with tweezers or by hand.



2. Perform your standard staining protocol by sequentially dipping the slide in the respective solutions.



TIP – For mounting slide samples, a hardening permanent mounting medium is recommended, such as Fluoroshield™ (Sigma-Aldrich), Vectashield® (Vector Laboratories Inc.), or ProLong® Antifade (ThermoFisher Scientific).

ibidi Mounting Medium is not recommended, as it is non-hardening and remains liquid, which is advantageous for μ -Slides and μ -Dishes but not suitable for mounting slide samples.

Chemical Compatibility

The following table provides some basic information on the chemical and solvent compatibility of the 12 Well Chamber, removable. For a full list of compatible solvents and more information on chemical compatibility, visit [ibidi.com/chemicals](https://www.ibidi.com/chemicals).

Chemical / Solvent	Compatibility
Methanol	Yes
Ethanol	Yes
Formaldehyde	Yes
Acetone	Yes, without lid
Immersion oil	Yes

Mounting

Mount the slide using a 24 × 60 mm cover-slip (ibidi #10811) and a permanent mounting medium of your choice.

For research use only!

Further information can be found at [ibidi.com](https://www.ibidi.com). For questions and suggestions, please contact us by e-mail at info@ibidi.com or by telephone at +49 (0)89/520 4617 0.
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