

The 12 well Chamber, removable is a removable silicone chamber for cell culture and immunofluorescence stainings. It allows the use of standard cultivation, staining and mounting techniques with coverslip sealing. 12 well Chamber, removable supports upright microscopy and long-term storage of cell culture microscopy samples after mounting the glass slide with a coverslip¹. 12 well Chamber, removable is not recommended for live cell microscopy on inverted microscopes since cells grow on a 1 mm microscopy glass slide.

¹Suitable 24 mm × 60 mm coverslips are provided by ibidi (10811).

Material

The 12 well Chamber, removable is comprised of a self-adhesive 12 well silicone gasket mounted on a standard microscopy glass slide. The gasket is manufactured from biocompatible silicone material. Although both materials are autoclavable and compatible with alcohols, we do not recommend reusing them.

Geometry

12 well Chamber, removable provides a standard slide format according to ISO 8037/1.

Geometry of the 12 well Chamber, removable

| | |
|--------------------------------------|--|
| Number of wells | 12 |
| Dimension of wells (w × l × h) in mm | 7.5 × 7.5 × 8 |
| Volume per well | 250 µl |
| Total height with lid | 11 mm |
| Growth area per well | 0.56 cm ² |
| Coating area per well | 1.9 cm ² |
| Bottom material, size in mm | standard glass slide 26 × 76 × 1, ground edges, twin frosted ends 13 mm |

Surfaces and Coatings

12 well Chamber, removable is mounted on an uncoated glass slide with twin frosted ends. Specific coatings on glass are possible following this protocol:

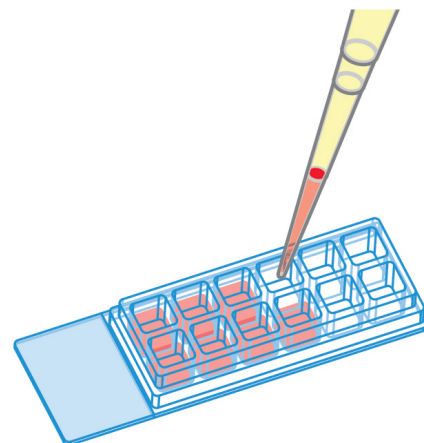
- Prepare your coating solution according to the manufacturer's specifications or reference.
- Apply 250 µl per well and leave at room temperature for at least 30 minutes.
- Aspirate the solution and wash with ultra-pure water. Let dry at room temperature.

Seeding Cells

Trypsinize and count cells as usual. Dilute the cell suspension to the desired concentration. 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.

- Apply 250 µl cell suspension into each well. Avoid shaking as this will result in inhomogeneous distribution of the cells.
- Cover wells with the supplied lid. Incubate at 37°C and 5% CO₂ as usual.

Undemanding cells can be left in their seeding medium for up to three days and grow to confluence there. However, best results might be achieved when the medium is changed every 1–2 days. Carefully aspirate the old medium and replace it by 250 µl/well fresh medium.



Solvents for Fixation, Staining and Other Purposes

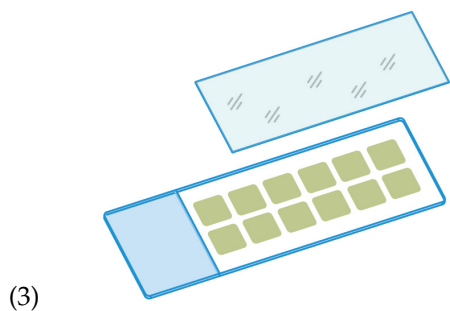
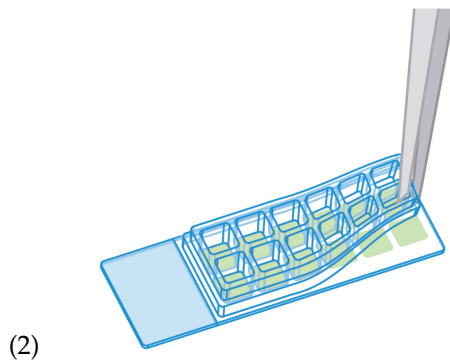
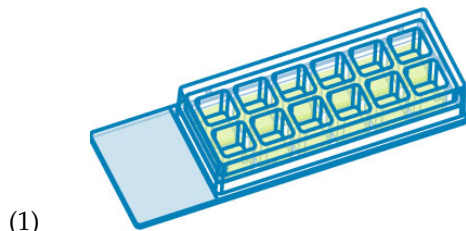
12 well Chamber, removable is compatible to methanol, acetone, acids, alkalis, PFA, DMSO, silicone oil, and mineral oil for cell culture.

Immunofluorescence Microscopy

After cultivation, cells can be fixed and stained before or after removing the silicone gasket.

Single well technique:

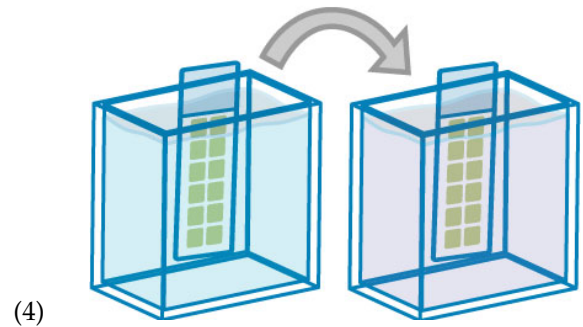
- Carry out all necessary steps (fixation, permeabilization, staining, washing) in single wells (1).
- Starting from one edge, remove the silicone gasket by hand or tweezers (2).
- Mount slide with a permanent mounting medium of your choice and a 24 mm × 60 mm coverslip (10811) (3).



Parallel technique:

- Starting from one edge, remove the silicone gasket by hand or tweezers (2).
- Carry out all necessary steps (fixation, permeabilization, staining, washing) dipping the slide into the solutions. A coverslip (24 mm × 60 mm) can be used during staining for reducing the volume (4).

- Mount slide and coverslip with a permanent mounting medium of your choice and a coverslip 24 mm × 60 mm (10811) (3).



ibidi Mounting Medium is not recommended for 12 well Chamber, removable because it is non-hardening and stays a liquid (which is advantageous for μ -Slides and μ -Dishes).

Tips:

The day before seeding the cells we recommend placing the cell medium and slides into the incubator for equilibration. This will prevent the liquid inside the slide or channel from creating air bubbles over the incubation time.

For mounting of slide samples, a permanent mounting medium is recommended. ibidi Mounting Medium is not recommended because it is non-hardening and stays a liquid (which is advantageous for μ -Slides and μ -Dishes).

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-Lysine | 18 months |
| Fibronectin | 4 months |

12 well Chamber, removable Family



| Cat. No. | Description | Characteristics |
|----------|--|-----------------|
| 81201 | 12 well Chamber, removable: microscopy glass slide | sterilized |
| 10811 | Coverslips for 12 well Chamber, removable, # 1.5H ($170 \pm 5 \mu\text{m}$) D263 M Schott glass, 24 mm × 60 mm | unsterile |

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.

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