

## Collagen Cell Carrier

### Before starting please note

When working with CCC use appropriate cell culture plastics, media and reagents as well as aseptic techniques and ensure adequate growth environments.

All liquids should be pre-warmed at least to room temperature. Always add liquids into the well along the sidewall.

After attachment to the bottom of the well plate avoid touching the CCC membrane until its translocation if intended.

After detachment of the CCC the whole, intact cell-scaffold complex can be removed and transferred.



*Collagen Cell Carrier (CCC) is intended for research use only. It is neither intended for human nor animal diagnostic, therapeutic use or any other clinical uses!*

### Corresponding documents:

- Product Data Sheet - Collagen Cell Carrier (CCC)
- Application Note - Detachment of cells cultured on fibrous collagen surfaces
- Application Note - Staining of cells grown on fibrous collagen surfaces with the life cell tracking dye BDTM DiIC12(3)

All data and recommendations correspond to the present state of our knowledge; they are published without engagement. We reserve the right to make alterations and additions in line with technical developments without prior notice. The customer is obliged to check whether our products meet with his own technical requirements. We shall be glad to answer any queries.

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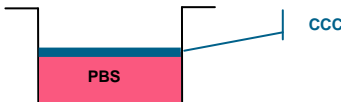
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### Attachment of the CCC to the well bottom and cell seeding

	Ø 34 mm (6-well)	Ø 21 mm (12-well)	Ø 14 mm (24-well)	Ø 10 mm (48-well)	Ø 7 mm (96-well)	Ø 88 mm (10 cm dishes)
Volume of PBS	1000 µL	500 µL	250 µL	150 µL	50 µL	6 mL

- Preload each well with the above given volume of PBS (pH7.3 w/o Ca<sup>2+</sup> / Mg<sup>2+</sup>)  
**Important: Do not exceed these volumes! For cell types known to be sensitive to phosphate buffers, the CCC may be rehydrated with ddH<sub>2</sub>O before incubation with medium.**
  - Take the CCC out of the blister by using a pair of sterile forceps and place it on top of the liquid.  
**Important: Do not submerge!**
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- Incubate for 30 min at room temperature.
  - Carefully aspirate the remaining PBS.  
Assure that the CCC is flatly, without bubbles positioned on the bottom of the well.
  - Important: Dry open under the operating laminar flow hood overnight!**  
The dry CCC is slightly opaque and will become transparent again after wetting.
  - Prior to cell seeding, the CCC needs to be equilibrated (to reach physiological pH) with the appropriate volume of pre-warmed medium for at least 30 min at 37°C in the CO<sub>2</sub> incubator.
  - Change the medium just prior to seeding cells on the CCC.

### Detachment of cell loaded CCC from the well plate

- Aspirate the medium from the well except a small volume. Wet the inner sidewall of the well with a small amount of medium.
- Grab the cell-scaffold complex at one side with a pair of forceps and remove it from the bottom of the well by pulling gently to the opposite well side in a steep angle. Optionally, to loosen the CCC, circuit it once with the tip of a pair of pointed forceps. In case it sticks rather tightly, remove small areas of the CCC boundary at several sides before pulling.
- Instantly, place a glass slide or a similar support close to the well, wet it with any buffer, water or medium. Carefully pull the CCC directly from the sidewall on top of the support.  
Avoid collapsing of the CCC.