# Instructions for use

## Thermo Scientific Nunc UpCell Surface Temperature-Responsive Cell Culture Surface

Thank you for purchasing a product with Thermo Scientific Nunc UpCell Surface.

Please ensure that there is no damage to the package and read the following instructions carefully to ensure the maximum performance of this product. The product is intended for research purposes only. Any other use is not warranted by Thermo Fisher Scientific. Do not use the product for clinical or diagnostic purposes.

Products with Nunc UpCell™ Surface are intended for single-use only. Do not re-use the product as cell-secreted and medium-derived components may be adsorbed to the surface after cell harvest, compromising performance in repeated use. Re-sterilization may compromise the Nunc UpCell Surface properties.

### **Storage**

The product should be stored at room temperature (20–25°C) and out of direct sunlight.

#### In vitro cell culture

- The temperature-responsive polymer covalently immobilized to the polystyrene surface of each culture area becomes slightly hydrophobic above 32°C, allowing cells to adhere. It is recommended that cells are incubated above 32°C and preferable at 37°C.
- When seeding cells, use pre-warmed (37°C) medium to ease attachment of cells. Seed cells evenly over the culture area to avoid uneven distribution of the cells in culture. Avoid scratching the surface with pipette tips, as this can destroy the temperature-responsive surface.
- Attachment time for cells may vary depending on cell type.
- Should you have difficulty in attaching cells, pre-coating of the culture surface with collagen, fibronectin, poly-lysine, laminin or the use of medium with serum may improve cell attachment.
- The Nunc UpCell Surface becomes hydrophilic below 32°C, prompting cell detachment from the surface.
- To avoid undesired cell detachment during medium changes, pre-warm culture medium to 37°C. Observations under a microscope should also be kept short in order to prevent cooling and undesired cell detachment.

### **Detachment of cells into suspension**

- 1. Culture cells on the Nunc UpCell Surface to a density of less than 50–70% confluence (when cell-cell junctions are not established, harvested cells are readily separated by slight agitation and pipetting).
- 2. Place the product with Nunc UpCell Surface in an incubator set at 20°C in order to detach the cells. Alternatively, the product may be placed at room temperature (20–25°C) under sterile conditions. For quicker cell detachment, the culture medium can be exchanged to 20–25°C culture medium prior to detachment.
- **3.** Cells should detach spontaneously within 40 minutes, depending on the cell type and detachment conditions:

Conditions	20°C incubator	Room temperature (20–25°C)
Incubation time	10–30 minutes	15–40 minutes

**4.** Cells that fail to detach spontaneously can be detached by gentle plate/dish agitation, or by gently flushing the surface with medium.

# Detachment and transfer of cell sheet (For additional information, go to www.nuncbrand.com/go/upcell)

Only applicable to Nunc UpCell 3.5 cm Dishes and Multidish 6, Cat. Nos. 174904 and 174901, respectively.

A contiguous cell layer can be detached from the Nunc UpCell Surface using the supplied membrane:

- 1. Culture cells on the Nunc UpCell Surface to a confluent layer.
- 2. Aspirate all medium and add fresh medium (approximately 50 µl per dish or well) to prevent the cells from drying out. Gently place the membrane on top of the cell layer. Avoid creases or air bubbles between the membrane and the cell layer.
- 3. Place the Nunc UpCell Surface product at 20–25°C for 5–6 minutes (detachment time may vary depending on cell type).
- **4.** Use sterile forceps and gently detach the cell layer around the rim of the membrane. Grasp under the membrane and cell layer with the forceps and carefully withdraw from the **Nunc UpCell Surface**.
- 5. Transfer the membrane with the attached cell layer facing downwards to the desired surface (bare cultureware or another cell layer). Add fresh medium (approximately 10 –25 µl/cm² dish or well) and leave it undisturbed at 37°C for at least 30 minutes to allow for direct attachment of the cell layer to the new surface. Attachment to bare cultureware usually takes longer time than to another cell layer.
- **6.** Add 1 ml of medium on top of the membrane and gently withdraw the membrane from the cell layer, using the forceps.

The product is shipped under strict quality control. However, should you have any claims or questions, please do not hesitate to contact us.

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