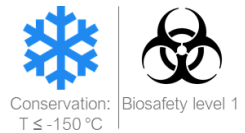


# Neonatal Human Epidermal Keratinocytes

Product sheet, catalog n° CTICC.1.2.2

## General Information

- **Organism:** Human (*Homo sapiens*)
- **Tissue:** Skin
- **Cell Type:** Skin keratinocytes from single donor
- **Location:** Foreskin or other part (see Certificate of Analysis)
- **Gender:** Male or Female (see Certificate of Analysis)
- **Age:** 0-1Y (see Certificate of Analysis)
- **Phototype:** (see Certificate of Analysis)



## Cell Characteristics

- **Cell properties:** Adherent
- **Morphology:** Polygonal Cobblestone-shaped
- **Isolation:** Enzymatic dissociation
- **Cell passage:** P0 to P2 (see Certificate of Analysis)
- **Minimum number of population doublings:** 10
- **Cell viability:** Minimum 70% viability when thawed from cryopreservation
- **Cell conditioning:** Supplied as vials of 1M cells
- **Cryopreservation medium:** Frozen with 90% serum-free cryopreservation medium + 10% DMSO
- **Storage condition:** Liquid nitrogen
- **Batch specific information:** (see Certificate of Analysis)

## Safety and Quality Control

- **Biosafety level:** 1
- **Contamination:** Use mandatory laboratory protections and handle with care tissues and cells derived from human samples to avoid any contamination of the operator
- **Viral testing:** negative for HIV, HBV, HCV
- **Sterility testing:** Negative for mycoplasma, bacteria and yeasts

## Handling upon delivery and storage

- Check that the containers are intact and free of damage
- If cells are not used immediately, place the vials at -150°C or below upon delivery

## Growth medium

- **Recommended medium reference:** CTIGM.Kerat: Growth Medium for Keratinocytes

## Thawing and culturing procedure for frozen cells

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- 1 - Add 0.12 ml per cm<sup>2</sup> of medium to the culture vessel (Recommended: 9mL in T75 flask)
- 2 - Add 13 ml of PBS solution to a 15 ml conical tube, and warm in a water bath to 37 °C
- 3 - Thaw cryovial by swirling in a water bath at 37 °C. As soon as the content has thawed, start step 4
- 4 - Once thawed, add the cell suspension to the warmed PBS in sterile conditions
- 5 - Spin the tube at 250 g for 7 minutes to pellet the cells
- 6 - Resuspend the cells in the appropriate volume of recommended medium
- 7 - Seed the cells in the culture vessel at a concentration of 5 000 to 10 000 cells per cm<sup>2</sup>
- 7 - Incubate at 37°C, 5% CO<sub>2</sub> atmosphere, 95% humidity
- 8 - After 24 hours of incubation, change the medium to remove any debris
- 9 - Continue to incubate and change the medium every 3-4 days

## Subculturing

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- 1- Start subculturing when cells reached 80%-90% confluence
- 2 - Preheat TrypLE (non-toxic for cell - trypsin substitute) and recommended medium.
- 3 - Remove the medium from the flask
- 4 - Wash the cells quickly with PBS without Ca<sup>2+</sup> Mg<sup>2+</sup>
- 5 - Add 0.06 mL per cm<sup>2</sup> of TrypLE for 10-12 min at 37 °C in the incubator
- 6 - Remove the cells from the flask by pipetting several times and wash the flask with recommended medium for remaining cells
- 7 - Centrifuge the cells in recommended medium at 250 g for 7 minutes to pellet the cells
- 8 - Remove the supernatant and resuspend the pellet in recommended medium
- 9 - Seed the cells in the culture vessel at a concentration of 5 000 cells per cm<sup>2</sup>
- 10 - Incubate at 37 °C, 5% CO<sub>2</sub> atmosphere, 95% humidity
- 11 - After 24 hours of incubation, change half of the medium to remove any debris
- 12 - Continue to incubate and change the medium every 3-4 days

## Associated products

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- **CTICC.1.1.2:** Neonatal Human Dermal Fibroblasts, Cryopreserved, 10<sup>6</sup> cells
- **CTICC.1.3.2:** Neonatal Human Melanocytes, Cryopreserved, 10<sup>6</sup> cells
- **SKIN BIOPSIES:** Fresh, Flash Frozen, FFPE, OCT-embedded

## Provisions

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- Cells and tissues are intended for **research use only** and shall not be used for human trials, animal trials, or diagnostics.
- **Consent:** the original tissues have been obtained after informed consent of the patient under the provisions required by French Law.
- **Primary Human cells** are not immortalised cell lines and may not be continually subcultured.