## Human Amniotic Membrane

 Mesenchymal Stem CellsProduct sheet, catalog $\mathrm{n}^{\circ}$ CTICC1. 12

## General Information

- Organism: Human (Homo sapiens)
- Tissue: Amniotic membrane
- Cell Type: Amniotic membrane mesenchymal stem cells from single donor
- Location: Stem cell extracted from the amniotic membrane
- Gender: Male or Female (see Certificate of Analysis)
- Age: Newborn (see Certificate of Analysis)
- Phototype (Fitzpatrick scale): (see Certificate of Analysis)


## Cell Characteristics

- Cell properties: Adherent
- Morphology: Bipolar, spindleshaped, fibroblast-like morphology
- Isolation: Enzymatic dissociation
- Cell passage: P0 to P2 (see Certificate of Analysis)
- Minimum number of population doublings: 15
- Cell viability: Minimum $80 \%$ viability when thawed from cryopreservation
- Cell conditioning: Supplied as vials of $1.10^{6}$ cells
- Cryopreservation medium: Frozen with $90 \%$ serum-free cryopreservation medium + 10\% DMSO
- Storage condition: Liquid nitrogen
- Batch specific information: Included in the 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 $-150^{\circ} \mathrm{C}$ or below upon delivery


## Thawing and culturing procedure for frozen cells

1 - Add $0,12 \mathrm{ml}$ per $\mathrm{cm}^{2}$ of medium to the culture vessel
2 - Add 13 ml of PBS solution to a 15 ml conical tube, and warm in a water bath to $37^{\circ} \mathrm{C}$
3 - Thaw cryovial by swirling in a water bath at $37^{\circ} \mathrm{C}$. 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 5000 to 10000 cells per $\mathrm{cm}^{2}$
8 - Incubate at $37^{\circ} \mathrm{C}, 5 \% \mathrm{CO}_{2}$ atmosphere, $95 \%$ humidity
9 - After 24 hours of incubation, change the medium to remove any debris
10 - Continue to incubate and change the medium every 3-4 days.

## Subculturing

1- Start subculturing when cells reached $70 \%-80 \%$ 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 Ca2+ Mg2+
5 - Add $0,06 \mathrm{~mL}$ per $\mathrm{cm}^{2}$ of TrypLE for $5-15 \mathrm{~min}$ at $37^{\circ} \mathrm{C}$ 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 5000 cells per $\mathrm{cm}^{2}$
10 - Incubate at $37^{\circ} \mathrm{C}, 5 \% \mathrm{CO}_{2}$ atmosphere, $95 \%$ humidity
11 - After 24 hours of incubation, change half of the medium to remove any debris
12 - Continue to incubate and change half of the medium every 3-4 days.

## Associated products

- CTICC.1.11: Human Amniotic Membrane Epithelial Stem Cells, Cryopreserved, $10^{6}$ cells
- CTIINT.1.5: Human Amniotic Membrane, Fresh, $1 \mathrm{~cm}^{2}$
- CTIINT.1.1: Human Placenta, Full Thickness, Fresh, $1 \mathrm{~cm}^{3}$
- CTIINT.1.9: Human Umbilical Cord, Fresh, 1 cm


## Provisions

- 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.

