Human Dermal Fibroblasts (Neonatal)

ORDER INFORMATION

Name of Cells: Human Dermal Fibroblasts (HDFCs-neo)
Catalogue Number: cAP-0008
Product Format: Proliferating cells in T25 or a Frozen Vial
Cell Number: > 90% confluent in T25 flask or Frozen Vial (>5x 10^5/vial)

General Information
HDFCs-neo are isolated from normal neonatal foreskin tissue samples and shipped in proliferating culture with >90 confluence (the cells are provided @ passage 1) or frozen vial (>5x10^5 cells). DMEM contains 5% Fetal Bovine Serum (Full medium) is recommended for cell culture and these cells have an average minimum population doubling levels > 14 when cultured following the detailed protocol described below). HDFCs-neo are tested negative for HIV-1, HBV, HCV, and mycoplasma.

Product Use: HDFCs-neo are for research use only.

Shipping: Proliferating culture in T25 flask or a Frozen Vial.

Handling of Arriving Cells

(1) When you receive the cells in a T25 flask, leave the T25 flask in 37°C CO2 incubator for 1 hour first, and then replace the transport medium with fresh full medium. Let the cells grow for 24 hour before subculture.

(2) When you receive the cells in a frozen vial, you can transfer the vial of cells into a -80°C freezer for short term storage or a liquid nitrogen tank for long term storage. Thaw the cells in a 37°C water bath, and then transfer the cells into a T25 flask pre-coated with Quick coating solution (cAP-01) as described in details in Subculture Protocol.

Subculture Protocol

A) Pre-coating of T25 flasks: Add 2ml of Quick Coating Solution (cAP-01) into one T25 flask and make sure whole surface of the flask is covered with the coating solution. Five minutes later, dispose excessive Quick Coating Solution by aspiration and the flask is ready to be used (no need for overnight incubation when using Quick Coating Solution). Other extracellular matrix can be used including gelatin, collagen, and fibronectin and you are advised to test the conditions for using those materials in advance.

B) Rinse the cells in T25 flask with 5ml HBSS (Room Temperature, RT) twice.

C) Add 2ml of Trypsin/EDTA (RT) (cAP-23) into one T25 flask (make sure the whole surface of the T25 flask is covered with Trypsin/EDTA), and gently dispose the excessive Trypsin/EDTA solution within 20 seconds with aspiration.
D) Leave the T25 flask with the cells at RT for 1 minute (the cells usually will detach from the surface within 1-2 minutes). You can monitor the cells under microscope and when most of cells become rounded up, hit the flask against the bench surface, and the cells will move on the surface of the flask when monitoring under microscope.

E) Add 5ml Trypsin Neutralization Buffer (cAP-28) and spin the cells down with 800g for 5 minutes.

F) Re-suspend the cell pellet with 10 - 20ml of full medium and the cell suspension is transferred directly into 2 or 4 pre-coated T25 flasks (5ml each, and the cells are sub-cultured at 1:2 to 1:4 ratios)

G) Change medium every 2-3 days and cells usually become confluent within 7 days (when split at a 1:4 ratio).

**Related products**

<table>
<thead>
<tr>
<th>Product Name</th>
<th>Code</th>
<th>Volume</th>
<th>Origin</th>
</tr>
</thead>
<tbody>
<tr>
<td>Quick Coating Solution</td>
<td>cAP-01</td>
<td>240ml</td>
<td>Angio-Proteomie</td>
</tr>
<tr>
<td>Endothelial Growth Medium</td>
<td>cAP-02</td>
<td>500ml</td>
<td>Angio-Proteomie</td>
</tr>
<tr>
<td>Endothelial Basal Medium</td>
<td>cAP-03</td>
<td>500ml</td>
<td>Angio-Proteomie</td>
</tr>
<tr>
<td>HBSS w/o Ca(^{2+}), Mg(^{2+})</td>
<td>cAP-11</td>
<td>100ml</td>
<td>Angio-Proteomie</td>
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<tr>
<td>Trypsin/EDTA Solution</td>
<td>cAP-23</td>
<td>100ml</td>
<td>Angio-Proteomie</td>
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<tr>
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<td>100ml</td>
<td>Angio-Proteomie</td>
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</tbody>
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Caution: Handling human tissue derived products is potentially bio-hazardous. Although each cell strain is tested negative for HIV, HBV and HCV DNA, diagnostic tests are not necessarily 100% accurate; therefore, proper precautions must be taken to avoid inadvertent exposure. Always wear gloves and safety glasses when working with these materials. Never mouth pipette. We recommend following the universal procedures for handling products of human origin as the minimum precaution against contamination.