NEUROMICS

Human Dermal Fibroblast Cells

Catalog #: HDF001

Cell #: >5x10^5 cells

Storage: Liquid Nitrogen until ready for culture.
While culturing keep in 37°C CO_2 incubator (95% air, 5% CO_2)

Product Format: Frozen Vial

GENERAL INFORMATION

Human cells exhibiting fibroblast morphology isolated from normal dermal tissue. Cell populations are offered at passage 1 in a frozen vial. It is recommended to culture these cells following the protocols described below.

Product is for Research use only.

Frozen Vials are shipped in a Dry Ice Package.

STATEMENT

Handling human tissue derived products is potentially bio-hazardous, despite testing negative for HIV, HBV, and HCV DNA. Nonetheless, proper precautions must be taken to avoid inadvertent exposure.

SPECIAL NOTES:

- We strongly advise our customers to use medium and related products recommended by Neuromics, because our cells were grown and adapted using our products.
- The growth featured of our cells cannot be guaranteed if the specific growth mediums stated in our datasheets are not used.
- Due to the sensitive nature of primary cells and cell lines, all quality related issues about the cells must be reported back to us within one month after receiving the products. Cells will not be replaced after the one-month period.

UNPACKING AND STORAGE INSTRUCTIONS

1. Check all containers for leakage or breakage. Remove the frozen cells from the dry ice packaging and immediately place the cells at a temperature below -130°C, preferably in liquid nitrogen vapor, until ready for use.

2. Complete medium: The base medium for this cell line is Stem Cell Complete Media (CAFM02).

HANDLING PROCEDURE

To ensure the highest level of viability, thaw the vial and initiate the culture as soon as possible upon receipt. If upon arrival, continued storage of the frozen culture is necessary, it should be stored in liquid nitrogen vapor phase and not at -70°C. Storage at -70°C will result in loss of viability.

1. Thaw the vial by gentle agitation in a 37°C water bath. To reduce the possibility of contamination, keep the O-ring and cap out of the water. Thawing should be rapid (approximately 2 minutes).

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2. Remove the vial from the water bath as soon as the contents are thawed. Decontaminate by dipping in or spraying with 70% ethanol. All the operations from this point on should be carried out under strict aseptic conditions.

3. It is recommended that the cryoprotective agent be removed immediately. Centrifuge the cell suspension at approximately 125 x g for 5 to 10 minutes. Discard the supernatant and resuspend the cell pellet in an appropriate amount of fresh growth medium.

4. Add 6.0 to 8.0 mL of AlphaBioCoat (cat. AC001) to the T-Flask for 15 minutes. Aspirate the solution after 15 minutes, rinse with 8ml of 1XPBS. Discard the 1XPBS. Transfer the cells to an appropriate size T-Flask.

5. It is important to avoid excessive alkalinity of the medium during recovery of the cells. It is suggested that, prior to the addition of the vial contents, the culture vessel containing the growth medium be placed into the incubator for at least 15 minutes to allow the medium to reach its normal pH (7.0 to 7.6).

6. Incubate the culture at 37°C in a suitable incubator. A 5% CO2 in air atmosphere is recommended if using the medium described on this product sheet.

PROTOCOL FOR THAWING THE CELLS AND SUBCULTURE

Note: If you have any questions or need clarification regarding the protocol for culturing these cells, please reach out to Dr. Jensen Auguste at (978) 608-1766 with your questions before beginning.

Note: Volumes used in this protocol are for 75 cm^2 flasks; proportionally reduce or increase amount of dissociation medium for culture vessels of other sizes.

1. Remove and discard culture medium. Briefly rinse the cell layer 0.25% trypsin, 0.53 mM EDTA solution. Remove the solution.

2. Add an additional 1 to 2 mL of Cell Detachment (ADF001) solution. Allow the flask to sit at room temperature (or at 37°C) until the cells detach. Add fresh culture medium, aspirate, and dispense into new culture flasks.

Subcultivation Ratio: A subcultivation ratio of 1:2 to 1:3 is recommended.

Medium Renewal: Every 2 to 3 days

Note: Growth of the cells is enhanced by the addition of tumor necrosis factor-alpha (TNF alpha) to the medium. Reagents for cryopreservation: Complete growth medium supplemented with 5% (v/v) DMSO.

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