

## HFF-1 Cells | 305790

## General information

## Description

HFF-1 is a human foreskin fibroblast cell line frequently used as a feeder layer for the culture of human embryonic stem cells (hESCs) and induced pluripotent stem cells (iPSCs). Derived from neonatal dermal tissue, HFF-1 cells provide essential extracellular matrix components and secrete key signaling molecules that promote hESC attachment and partially support their pluripotent state. These fibroblasts have been evaluated for their expression of several pluripotency-supporting growth factors, including TGF $\beta$ 1, activin A, and fibroblast growth factor 2 (FGF-2), although their effectiveness as feeder cells can vary depending on the specific line and culture conditions.

In comparative studies, human foreskin fibroblasts like HFF-1 secrete detectable levels of FGF-2 and activin A, though their secretion levels are generally lower than those observed in mouse embryonic fibroblasts. HFF-1 cells also express BMP-4 mRNA and protein, although secreted levels of BMP-4 dimers are extremely low and often undetectable in conditioned media, likely due to intracellular sequestration or inhibition by gremlin. Importantly, the secretion of growth factors by HFF-1 is modulated by mitotic inactivation (e.g., mitomycin C treatment) and media composition (e.g., KnockOut Serum Replacement vs. fetal bovine serum). The ability of HFF-1 cells to support undifferentiated hESC growth correlates with their secretion of activin A and TGF $\beta$ 1, although supplementation with exogenous activin A can improve the maintenance of pluripotency markers such as SSEA3 when these cells are used as feeders.

Overall, HFF-1 serves as a useful human-derived feeder cell model for stem cell culture systems aiming to reduce xeno-components. However, their capacity to maintain long-term undifferentiated hESC cultures is generally considered less robust than that of mouse-derived feeder cells unless combined with specific growth factor supplementation. Their human origin, however, makes them particularly attractive for clinical and translational stem cell applications where xeno-free conditions are essential.

**Organism** Human

**Tissue** Foreskin, skin

**Synonyms** HFF1

## Characteristics

**Age** 1 month

**Gender** Male

**Morphology** Fibroblast

**Cell type** Fibroblast of foreskin

**Growth properties** Adherent

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## Regulatory Data

<b>Citation</b>	HFF-1 (Cytion catalog number 305790)
<b>Biosafety level</b>	1
<b>NCBI_TaxID</b>	9606
<b>CellosaurusAccession</b>	CVCL_3285

## Biomolecular Data

<b>Mutational profile</b>	
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## Handling

<b>Culture Medium</b>	DMEM, w: 4.5 g/L Glucose, w: 4 mM L-Glutamine, w: 3.7 g/L NaHCO <sub>3</sub> , w: 1.0 mM Sodium pyruvate (Cytion article number 820300a)
<b>Supplements</b>	Supplement the medium with 15% FBS
<b>Dissociation Reagent</b>	Accutase
<b>Fluid renewal</b>	2 to 3 times per week
<b>Freeze medium</b>	As a cryopreservation medium, we use complete growth medium (including FBS) + 10% DMSO for adequate post-thaw viability, or CM-1 (Cytion catalog number 800100), which includes optimized osmoprotectants and metabolic stabilizers to enhance recovery and reduce cryo-induced stress.

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### Thawing and Culturing Cells

1. Confirm that the vial remains deeply frozen upon delivery, as cells are shipped on dry ice to maintain optimal temperatures during transit.
2. Upon receipt, either store the cryovial immediately at temperatures below  $-150^{\circ}\text{C}$  to ensure the preservation of cellular integrity, or proceed to step 3 if immediate culturing is required.
3. For immediate culturing, swiftly thaw the vial by immersing it in a  $37^{\circ}\text{C}$  water bath with clean water and an antimicrobial agent, agitating gently for 40-60 seconds until a small ice clump remains.
4. Perform all subsequent steps under sterile conditions in a flow hood, disinfecting the cryovial with 70% ethanol before opening.
5. Carefully open the disinfected vial and transfer the cell suspension into a 15 ml centrifuge tube containing 8 ml of room-temperature culture medium, mixing gently.
6. Centrifuge the mixture at  $300 \times g$  for 3 minutes to separate the cells and carefully discard the supernatant containing residual freezing medium.
7. Gently resuspend the cell pellet in 10 ml of fresh culture medium. For adherent cells, divide the suspension between two T25 culture flasks; for suspension cultures, transfer all the medium into one T25 flask to promote effective cell interaction and growth.
8. Adhere to established subculture protocols for continued growth and maintenance of the cell line, ensuring reliable experimental outcomes.

### Incubation Atmosphere

$37^{\circ}\text{C}$ , 5%  $\text{CO}_2$ , humidified atmosphere.

### Flask Coating

None

### Shipping Conditions

Cryopreserved cell lines are shipped on dry ice in validated, insulated packaging with sufficient refrigerant to maintain approximately  $-78^{\circ}\text{C}$  throughout transit. On receipt, inspect the container immediately and transfer vials without delay to appropriate storage.

### Storage Conditions

For long-term preservation, place vials in vapor-phase liquid nitrogen at about  $-150$  to  $-196^{\circ}\text{C}$ . Storage at  $-80^{\circ}\text{C}$  is acceptable only as a short interim step before transfer to liquid nitrogen.

## Quality Control & Molecular Analysis

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**Sterility**

Mycoplasma contamination is excluded using both PCR-based assays and luminescence-based mycoplasma detection methods.

To ensure there is no bacterial, fungal, or yeast contamination, cell cultures are subjected to daily visual inspections.