Product sheet

HROHep03 Cells | 300197

General information

Description	HROHep03 is a human hepatocellular adenocarcinoma cell line established from the primary liver tumor of a 71-year-old Caucasian female patient within the HRO biobank series of patient-derived tumor cell lines developed by PD Dr. Michael Linnebacher since 2006. The tumor was classified as a primary adenocarcinoma at TNM stage T0NxMx, grade 3, reflecting a high-grade hepatic adenocarcinoma without confirmed distant metastasis at the time of tissue harvest. HROHep03 grows as an adherent monolayer with fibroblast-like morphology and was confirmed free of human pathogenic viruses HBV, HCV, and HIV, consistent with the stringent quality control standards of the Linnebacher biobank series. The Cellosaurus accession is CVCL_2U72. HROHep03 is applicable in hepatocellular adenocarcinoma research, studies of high-grade liver tumor cell biology, drug sensitivity and resistance testing (sorafenib, cisplatin, 5-FU), liver tumor invasion and migration assays, and molecular pathway analysis. As part of the HRO biobank, this line provides a patient-specific biological resource that can be paired with matched immunological material from the same patient for personalized oncology research. Its fibroblast-like morphology distinguishes it phenotypically from the more common hepatocyte-like HCC lines and may reflect epithelial-to-mesenchymal features acquired during tumor progression or in vitro adaptation. HROHep03 is maintained as an adherent culture in DMEM:Ham's F12 (1:1) supplemented with 10% FBS at 37°C in a humidified 5% CO_2 atmosphere. Cells are subcultured with Accutase when approximately 80–90% confluent. Medium is renewed every 3–5 days; after thawing, allow at least 2 days for recovery before the first medium change.
Organism	Human
Tissue	Liver
Disease	Primary adenocarcinoma, T0NxMx stage, grade 3
Metastatic site	Not applicable (TNM stage T0NxMx; no confirmed distant metastasis at time of sample collection)
Applications	Hepatocellular adenocarcinoma research; high-grade HCC modeling; drug sensitivity testing (sorafenib, cisplatin, 5-FU); liver tumor invasion and migration; patient-matched HRO biobank studies

Characteristics

Age	71 years
Gender	Female
Ethnicity	Caucasian
Morphology	Fibroblast-like

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Cell type	Fibroblast-like (hepatocellular carcinoma)
Growth properties	Adherent

Regulatory Data

Citation	HROHep03 (Cytion catalog number 300197)
Biosafety level	1
NCBI_TaxID	9606
CellosaurusAccession	CVCL_2U72
GMO Status	No genetic modification; wildtype patient-derived hepatic adenocarcinoma cell line established by PD Dr. Linnebacher. Confirmed free of HBV, HCV, HIV.

Biomolecular Data

Viruses	Free of human pathogenic viruses HBV, HCV, HIV.

Handling

Culture Medium	DMEM:Ham's F12 (1:1), w: 3.1 g/L Glucose, w: 2.5 mM L-Glutamine, w: 15 mM HEPES, w: 0.5 mM Sodium pyruvate, w: 1.2 g/L NaHCO3 (Cytion article number 820400a)
Supplements	Supplement the medium with 10% FBS
Dissociation Reagent	Accutase
Doubling time	approx. 48 to 72 hours
Subculturing	Remove the old medium from the adherent cells and wash them with PBS that lacks calcium and magnesium. For T25 flasks, use 3-5 ml of PBS, and for T75 flasks, use 5-10 ml. Then, cover the cells completely with Accutase, using 1-2 ml for T25 flasks and 2.5 ml for T75 flasks. Let the cells incubate at room temperature for 8-10 minutes to detach them. After incubation, gently mix the cells with 10 ml of medium to resuspend them, then centrifuge at 300xg for 3 minutes. Discard the supernatant, resuspend the cells in fresh medium, and transfer them into new flasks that already contain fresh medium.
Split ratio	1 to 3

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Seeding density	2 x 10^4 cells/cm^2
Fluid renewal	Every 3 to 5 days
Post-Thaw Recovery	2 days
Freeze medium	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.
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°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°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 x 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°C, 5% CO_2, humidified atmosphere.

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Shipping Conditions	Cryopreserved cell lines are shipped on dry ice in validated, insulated packaging with sufficient refrigerant to maintain approximately −78 °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 °C. Storage at −80 °C is acceptable only as a short interim step before transfer to liquid nitrogen.

Quality Control & Molecular Analysis

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.