

ECV-304 Cells | 300452

General information

Description

The ECV-304 cell line is a spontaneously transformed cell line derived from human umbilical vein endothelial cells (HUVEC). Originally characterized and utilized as a model of endothelial cell biology, further genomic analysis revealed that ECV-304 cells were contaminated and are genetically identical to the T24 bladder carcinoma cell line. This revelation has significant implications for the interpretation of research data, particularly studies conducted under the assumption that ECV-304 was a true endothelial cell model.

Despite its endothelial mischaracterization, ECV-304 has been widely used in studies related to tumorigenesis, cytotoxicity, and drug screening, primarily due to its robust growth characteristics and the ease with which it can be cultured. The cells exhibit epithelial morphology and possess the ability to grow in a monolayer, making them a suitable in vitro model for studying various aspects of cancer biology, including cell proliferation, migration, and the cellular response to therapeutic agents. However, caution must be exercised in the interpretation of past studies where ECV-304 was used as a model of endothelial cells.

Given the genetic identification with the T24 cell line, researchers should consider ECV-304 as a bladder carcinoma model rather than an endothelial cell line. This understanding is crucial for the design and interpretation of experiments aimed at investigating cellular behaviors that are relevant to cancer research rather than endothelial cell function.

Organism Human

Tissue Bladder

Disease Carcinoma

Synonyms ECV 304, ECV304, ECV, E304, T24(ECV304)

Characteristics

Age 82 years

Gender Female

Morphology Epithelial-like

Growth properties Adherent

Regulatory Data

Citation ECV-304 (Cytion catalog number 300452)

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Biosafety level	1
NCBI_TaxID	9606
CellosaurusAccession	CVCL_2029

Biomolecular Data**Handling**

Culture Medium	Medium 199, w: 2.7 mM stable Glutamine, w: 2.2 g/L NaHCO ₃ , w: EBSS (Cytion article number 820101a)
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Supplements	Supplement the medium with 10% FBS
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Dissociation Reagent	Accutase
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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.
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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.
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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°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 $300 \times g$ 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°C , 5% CO_2 , humidified atmosphere.

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

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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.