

H4-II-E-C3 Cells | 305336**General information****Description**

The H4-II-E-C3 cell line is a well-established rat hepatoma cell line derived from liver carcinoma. This line is widely used in research focused on liver-specific functions and the metabolic processes that occur within hepatocytes. One notable aspect of H4-II-E-C3 cells is their high expression of the high-affinity glutamate transporter GLT-1A. This feature distinguishes them from normal rat hepatocytes, which do not exhibit significant levels of this transporter. The expression of GLT-1A in H4-II-E-C3 cells allows for the efficient study of glutamate metabolism and transport, providing insights into liver cell transformation and tumor cell metabolism.

Additionally, H4-II-E-C3 cells are used to study aldehyde metabolism, particularly the activity of mitochondrial class 2 aldehyde dehydrogenase (ALDH2). This enzyme is critical for metabolizing acetaldehyde produced during ethanol oxidation, making this cell line suitable for research on alcohol metabolism and its cellular effects. The use of an "acetaldehyde clamp" technique in these cells has allowed precise measurement of ALDH2 activity without the need for mitochondrial isolation, facilitating the study of enzyme kinetics and the impact of various inhibitors on aldehyde detoxification.

Organism

Rat

Tissue

Liver

Disease

Hepatocellular carcinoma

Synonyms

H4IIEC3, H4-IIE-C3, H4-II-EC3, H-II-E-C3

Characteristics**Breed/Subspecies**

AxC

Age

Unspecified

Gender

Male

Growth properties

Adherent

Regulatory Data**Citation**

H4-II-E-C3 (Cytion catalog number 305336)

Biosafety level

1

H4-II-E-C3 Cells | 305336**NCBI_TaxID** 10116**CellosaurusAccession** CVCL_0285**GMO Status** GMO-S1: This rat hepatocellular carcinoma cell line (H4-II-E-C3) contains undefined modifications classified as GMO, stably present in the transformed hepatic cell line. This classification applies only within Germany and may differ elsewhere.**Biomolecular Data****Protein expression** Gene expression: tyrosine aminotransferase, albumin, transferrin, prothrombin**Tumorigenic** Yes, in AxC rats**Handling****Culture Medium** DMEM, w: 4.5 g/L Glucose, w: 4 mM L-Glutamine, w: 3.7 g/L NaHCO₃, w: 1.0 mM Sodium pyruvate (Cytion article number 820300a)**Supplements** Supplement the medium with 10% FBS**Dissociation Reagent** Accutase**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.**Seeding density** $1-3 \times 10^4/\text{cm}^2$ **Fluid renewal** 2 to 3 times per week**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 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% CO₂, 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.