

**H-4-II-E Cells | 305204**

**General information**

**Description**

The H-4-II-E cell line is a hepatoma-derived cell line, specifically developed from the liver tissue of an adult rat. This cell line is frequently utilized in research focused on liver physiology and pathology, particularly in studies related to hepatocellular carcinoma and liver metabolism. The H-4-II-E cells are epithelial-like in morphology and are known for their rapid growth rate and high cell density capabilities, making them a valuable model for high-throughput screening and toxicology studies.

Notably, H-4-II-E cells exhibit several differentiated functions of hepatocytes, including albumin secretion, ammonia production, and the presence of cytochrome P450 enzymes. This makes them an excellent tool for studying drug metabolism, enzyme induction, and interactions that may occur in human liver disease. Moreover, these cells have been used to investigate mechanisms of liver regeneration and cancer progression, providing insights into cellular responses to various stimuli such as cytokines and growth factors.

**Organism**

Rat

**Tissue**

Liver

**Disease**

Rat hepatocellular carcinoma

**Synonyms**

H-4-II-E, H4IIE

**Characteristics**

**Breed/Subspecies**

AxC

**Gender**

Male

**Morphology**

Epithelial

**Growth properties**

Adherent

**Regulatory Data**

**Citation**

H-4-II-E (Cytion catalog number 305204)

**Biosafety level**

1

**NCBI\_TaxID**

10116

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CellosaurusAccession CVCL\_0284

**Biomolecular Data****Handling****Culture Medium** EMEM (MEM Eagle), w: 2 mM L-Glutamine, w: 2.2 g/L NaHCO<sub>3</sub>, w: EBSS (Cytion article number 820100a)**Supplements** Supplement the medium with 10% FBS and 1% NEAA**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** 2 to 4 x 10<sup>4</sup> cells/cm<sup>2</sup>**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  $-150^{\circ}\text{C}$  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.

### 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.