

**LMH Cells | 601411**

**General information**

**Description**

LMH cells, derived from a Leghorn male hepatoma, are a versatile cell line widely used in biological research. Tomoyuki Kitagawa established them in 1981 at the Cancer Institute in Tokyo, Japan. These cells have an epithelial phenotype and are particularly useful for studying host-pathogen interactions in the gastrointestinal tract of poultry.

LMH cells are adherent and exhibit a dendritic-like morphology. They express glucose-6-phosphatase and weak canalicular ATPase activity. With a triploid karyotype and six marker chromosomes, these cells display distinct genetic characteristics.

Notably, LMH cells have been shown to efficiently support duck hepatitis B virus (DHBV) DNA synthesis when transfected with viral constructs. This makes them an invaluable tool for virology research, particularly in the context of poultry-related viral infections.

The derivation of LMH cells involved inducing tumorous nodules in the liver of Leghorn chickens through long-term treatment with diethylnitrosamine. These cells have also been chemically transformed, allowing for their immortalization and continuous propagation in culture.

In terms of tumorigenicity, LMH cells have the ability to form tumors in athymic nude mice. This characteristic makes them an important model for studying hepatocellular carcinoma. LMH cells express the estrogen receptor and can be induced to express the liver-specific apolipoprotein II (apoII) gene. This indicates their involvement in estrogen signaling pathways and lipid metabolism. To culture LMH cells, it is necessary to precoat tissue culture vessels with collagen. This ensures proper cell adhesion and growth.

**Organism**      Chicken

**Tissue**          Liver

**Disease**        Hepatocellular carcinoma

**Applications**    The cell line is useful for transfection studies.

**Synonyms**        Leghorn Male Hepatoma cell line

**Characteristics**

**Breed/Subspecies**    Leghorn

**Age**                16 months

**Gender**            Male

**Morphology**        Epithelial-like, Dendritic like.

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**Growth properties**

Adherent. It may take a couple of days until cells grow in fully adherent colonies.

**Regulatory Data****Citation**

LMH (Cytion catalog number 601411)

**Biosafety level**

1

**NCBI\_TaxID**

9031

**CellosaurusAccession**

CVCL\_2580

**Biomolecular Data****Receptors expressed**

Estrogen (low level expression).

**Tumorigenic**

LMH cells form tumors in athymic mice.

**Products**

Glucose-6-phosphatase, canalicular ATPase activity (weak)

**Karyotype**

Triploid, modal number = 116, six marker chromosomes

**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**

LMH cells attach better to tissue culture vessels which have been precoated with Collagen. Remove medium and rinse the adherent cells using PBS without calcium and magnesium (3-5 ml PBS for T25, 5-10 ml for T75 cell culture flasks). Add Accutase (1-2 ml per T25, 2.5 ml per T75 cell culture flask), the cell sheet must be covered completely. Incubate at ambient temperature for 8-10 minutes. Carefully resuspend the cells with medium (10 ml), centrifuge for 3 min at 300 g, resuspend cells in fresh medium and dispense into new flasks which contain fresh medium.

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**Seeding density** 1 to  $3 \times 10^4$  cells/cm<sup>2</sup>

**Fluid renewal** Every 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<sub>2</sub>, 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.

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### 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.