LLC-MK2

305149

GENERAL INFORMATION

Description

LLC-MK2 is a continuous epithelial cell line established from the renal tissue of adult rhesus monkeys (*Macaca mulatta*). This cell line was originally isolated in the 1950s through the trypsinization of pooled kidney tissue from six rhesus monkeys. LLC-MK2 cells display adherent growth characteristics and have been extensively used in virology due to their high susceptibility to various viruses, including bovine viral diarrhea virus 1, human poliovirus 1, and human coxsackievirus B4. The cell line's origin and virus susceptibility make it an ideal model for studying viral replication and cytopathogenic effects.

The LLC-MK2 cell line is known for its ability to be cultured in chemically defined, serum-free media, which allows for controlled experimental conditions. Research has demonstrated that these cells can be adapted to serum-free conditions without compromising growth, although initial cultures were maintained in media containing significant amounts of horse serum. The adaptation to chemically defined media is particularly advantageous for virological studies, as it minimizes the variability introduced by serum and supports long-term cell line maintenance. Furthermore, the LLC-MK2 line has been shown to maintain virus sensitivity comparable to primary monkey kidney cells, making it a reliable tool for viral titration and vaccine production studies.

In addition to its role in virology, LLC-MK2 has also been investigated for its tumorigenic potential. Although it exhibits certain transformed characteristics, such as the ability to grow in soft agar, it does not form tumors in in vivo models, suggesting a limited tumorigenic risk. This characteristic further underscores its utility as a model cell line for in vitro studies, while confirming its unsuitability for therapeutic or in vivo applications.

Tissue	Kidney

Synonyms

- Llc-Mk2
- LLC-MK-2
- LLC-MK2 Original
- LLCMK2
- LLcMK2
- Lilly Laboratories Culture-Monkey Kidney 2

CLASSIFICATION

Organism	Rhesus macaque
Growth properties	Adherent
Biosafety level	1

CHARACTERISTICS

Morphology	Epithelial

IDENTIFIERS

Citation

LLC-MK2 (Cytion catalog number 305149)

NCBI TaxID	9544
Cellosaurus	CVCL_3009

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EXPRESSION AND SUSCEPTIBILITY

Protein expression

Plasminogen activator

HANDLING AND CULTURE

Culture medium

Medium 199, w: 2.7 mM stable Glutamine, w: 2.2 g/L NaHCO3, w: EBSS (Cytion article number 820101a)

Incubation atmosphere	37°C, 5% CO_2, humidified atmosphere.

Supplements

Supplement the medium with 1% horse serum

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	4 x 10^4 cells/cm^2
Fluid renewal	2 to 3 times per week

Thawing and culturing

Confirm that the vial remains deeply frozen upon delivery, as cells are shipped on dry ice to maintain optimal temperatures during transit.
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.
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.
Perform all subsequent steps under sterile conditions in a flow hood, disinfecting the cryovial with 70% ethanol before opening.
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.
Centrifuge the mixture at 300 x g for 3 minutes to separate the cells and carefully discard the supernatant containing residual freezing medium.
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.
Adhere to established subculture protocols for continued growth and maintenance of the cell line, ensuring reliable experimental outcomes.

QUALITY CONTROL AND CRYOPRESERVATION

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.

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.

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.

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.

DONOR

Donor age	Adult

LEGAL AND SAFETY

Safety precautions

When planning to store a cryovial in liquid nitrogen for future thawing, it is mandatory to adhere to stringent safety measures. Appropriate protective gloves and clothing are essential, and the use of a face mask or safety goggles is required during the transfer of frozen samples to or from the liquid nitrogen tank. This is to mitigate the risk of injury from potential cryovial explosions upon removal, which can result in the projection of sharp fragments.

Warranty

At Cytion, we stand behind the quality and viability of our products.
We guarantee that our cells will be viable at the start of culture for
60 days after shipment from our facilities.
We assure that all materials conform to the specifications described in the
Certificate of Analysis and/or Product Sheet.
For full details, please refer to our
Warranty Statement.

Material Transfer Agreement required	Yes