

A9 Cells | 305166

General information

Description

A9 cells are a fibroblast-like cell line derived from mouse adipose tissue. They were established as a subclone of the L929 parent strain by W. R. Earle in 1940. The parent strain was obtained from normal subcutaneous areolar and adipose tissue of a male C3H/An mouse.

A notable feature of these cells is that they express adenosine phosphoribosyl transferase (APRT) and hypoxanthine phosphoribosyl transferase (HPRT), denoted as APRT+ and HPRT+. These cells have been valuable in virus studies, particularly involving pseudorabies virus (PRV), vesicular stomatitis virus (VSV) of the Indiana strain, and herpes simplex virus (HSV).

A9 cells' sensitivity and response to these viruses have made them useful for studying viral replication, pathogenesis, and potential antiviral treatments. In immunology, A9 cells are used in various research areas. They are a valuable model for studying immune responses, antibody production, monoclonal antibody generation, and hybridoma technology.

Due to their rapid proliferation (doubling time of approximately 24 hours), A9 cells provide a sufficient cell supply for experiments and downstream applications. A9 cells have a fibroblast-like morphology and adhere to the culture substrate. Categorized as animal cells and belonging to the hybridoma cell type, A9 cells were formed by fusing B lymphocytes from *Mus musculus* (mouse) with myeloma cells from the same species.

This unique combination allows A9 cells to exhibit properties of both B lymphocytes and myeloma cells. Overall, A9 cells are a well-established fibroblast-like cell line utilized for studying viral infections, especially PRV, VSV, and HSV, and in immunology.

Organism Mouse

Tissue Subcutaneous Connective Tissue, Loose Connective Tissue And Fat, Normal

Synonyms A-9, A9 (Hamprecht), A9(Hamprecht), AG 9, GM00346, GM-346, GM346, GM00346B

Characteristics

Breed/Subspecies C3H/An

Age 100 days

Gender Male

Morphology Fibroblast-Like

Growth properties Adherent

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Regulatory Data

Citation	A9 (Cytion catalog number 305166)
Biosafety level	1
NCBI_TaxID	10090
CellosaurusAccession	CVCL_3984

Biomolecular Data

Antigen expression	H-2k
Tumorigenic	Yes, in nude mice.

Handling

Culture Medium	DMEM, w: 4.5 g/L Glucose, w: 4 mM L-Glutamine, w: 3.7 g/L NaHCO ₃ , 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.
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.

A9 Cells | 305166

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.

Flask Coating

None

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.