RAG

305190

GENERAL INFORMATION

Description

The RAG cell line is a nonreverting 8-azaguanine-resistant mutant derived from a renal adenocarcinoma of BALB/c mice. This line was developed through alternate animal-to-tissue culture passages to enrich for the tumorigenic population while eliminating normal stromal fibroblasts. RAG cells display an ameboid to epithelioid morphology with prominent cytoplasmic processes and are resistant to hypoxanthine-guanine phosphoribosyltransferase (HGPRT)-dependent selection methods due to their enzymatic deficiency. This resistance has facilitated their use in biochemical selection systems for somatic cell hybridization experiments.
RAG cells are widely utilized as a parental line in somatic cell fusion studies due to their compatibility with fusion procedures using inactivated Sendai virus. When fused with other cell lines, such as LM(TK-) or WI-38, the hybrids retain marker chromosomes and exhibit biochemical complementation of metabolic deficiencies. These hybrids have been instrumental in mapping genetic regulatory elements and studying gene expression, particularly in kidney-associated enzymes like the ES-2 esterase. RAG hybrids provide insights into both inter- and intraspecific chromosomal segregation and functional genomics.
In addition to their role in hybridization studies, RAG cells have served as a model to study the epigenetic regulation of gene expression. Hybrid cells involving RAG often show extinction and re-expression of specific genetic traits, depending on the retention or loss of particular chromosomes. This makes the RAG cell line a valuable tool in understanding the dynamics of genetic regulation and chromosomal stability in tumorigenic cells.

Tissue	Kidney
Disease	Mouse kidney carcinoma

Synonyms

- Rag

CLASSIFICATION

Organism	Mouse
Growth properties	Adherent
Biosafety level	1

CHARACTERISTICS

Morphology	Amoeboid

IDENTIFIERS

Citation

RAG (Cytion catalog number 305190)

NCBI TaxID	10090
Cellosaurus	CVCL_3575

EXPRESSION AND SUSCEPTIBILITY

Protein expression

Kidney specific esterase-2 (ES-2)

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HANDLING AND CULTURE

Culture medium

EMEM (MEM Eagle), w: 2 mM L-Glutamine, w: 2.2 g/L NaHCO3, w: EBSS (Cytion article number 820100a)

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

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.

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

Breed / subspecies	BALB/c

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

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