

SRA01-04 Cells | 305436

General information

Description

The SRA01/04 cell line is an immortalized human lens epithelial cell line developed to overcome the limitations associated with primary cultures of human lens epithelial (HLE) cells. These limitations include their low proliferative capacity and the difficulty of obtaining sufficient quantities of cells from donor lenses. SRA01/04 was derived from primary HLE cells obtained from infants undergoing surgery for retinopathy of prematurity. The immortalization was achieved by transfection with a plasmid carrying the SV40 large T antigen gene under the control of the RSV promoter. This approach avoids the use of live virus and permits selection of a uniform clone with consistent characteristics.

SRA01/04 cells exhibit stable proliferation over 130 passages with a population doubling level of approximately 3.5 per passage. Morphologically, they maintain a monolayer with epithelial characteristics. Chromosomal analysis revealed a hypotetraploid karyotype, consistent with an immortalized status. The isozyme phenotype confirmed their human origin. Importantly, the cell line retains lens-specific molecular features, including the expression of α A- and β B-crystallins and aldose reductase, though at lower protein levels than seen in primary HLE cultures. mRNA for both crystallins was also detected via RT-PCR, and sequencing confirmed 100% identity with published human sequences. This makes SRA01/04 the first established human lens cell line shown to express both α A- and β B-crystallins.

In addition to crystallin expression, SRA01/04 expresses aldose reductase, an enzyme involved in the polyol pathway, which is relevant to cataractogenesis. While optimal growth requires 20% fetal bovine serum, the cells can survive and proliferate with reduced serum levels or even in serum-free conditions if pre-attached to culture dishes. This adaptability, along with their stable lens-specific phenotype, renders SRA01/04 a valuable model for studying lens physiology, gene regulation in the lens epithelium, and mechanisms underlying cataract formation.

Organism Human

Tissue Eye, ocular lens, epithelium

Synonyms SRA 01-04, HLEC-SRA 01-04, HLE-SRA-01-04, SRA01-04 (HLE), SRA 01/04, HLEC-SRA 01/04, HLE-SRA-01/04, SRA01/04 (HLE)

Characteristics

Age 3 months

Gender Male

Morphology Epithelial-like

Cell type Ocular lense

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Growth properties	Adherent
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Regulatory Data

Citation	SRA01-04 (Cytion catalog number 305436)
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Biosafety level	1
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NCBI_TaxID	9606
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CellosaurusAccession	CVCL_7157
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GMO Status	GMO-S1: This SRA01-04 human lens epithelial line contains an SV40 large T-antigen construct enabling ocular lens research. This classification applies only within Germany and may differ elsewhere.
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Biomolecular Data

Protein expression	Genetic integration: Method=Transfection; Gene=UniProtKB; P03070; SV40 large T antigen
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Isoenzymes	LD, NP
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Handling

Culture Medium	DMEM low glucose
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Supplements	Supplement the medium with 10% FBS
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Dissociation Reagent	Accutase
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Seeding density	$2-4 \times 10^4/\text{cm}^2$
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Fluid renewal	2 times per week
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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°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% CO₂, 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.

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.