

## Calu-6 Cells | 300135

### General information

#### Description

The Calu-6 cell line is a human non-small cell lung carcinoma (NSCLC) cell line derived from the pleural effusion of a 61-year-old male patient. Established in 1975, this cell line has been a critical model in lung cancer research. Calu-6 cells exhibit a distinct epithelial morphology and have been used extensively to study the biology of lung cancer, including mechanisms of metastasis, drug resistance, and the tumor microenvironment. These cells are particularly noted for their ability to form tumors in xenograft models, which makes them highly valuable for in vivo studies of tumor growth and response to therapeutics.

Calu-6 is characterized by a high level of KRAS mutation, common in NSCLC, and provides a relevant model for studying the role of this oncogene in lung cancer. The cell line also displays several cytogenetic anomalies typical of cancer cells, such as complex karyotypes and aneuploidy, which contribute to its use in genetic studies. Research utilizing the Calu-6 cell line has helped in understanding the cellular mechanisms of lung cancer and in the development of therapeutic strategies. Its robust growth in culture and the ability to mimic clinical aspects of lung cancer make it an indispensable resource in oncological research.

**Organism** Human

**Tissue** Lung

**Disease** Adenocarcinoma

**Metastatic site** Pleural effusion

**Synonyms** CaLu-6, CALU-6, Calu.6, Calu 6, Calu6, CALU6, CaLu-06

### Characteristics

**Age** 61 years

**Gender** Female

**Ethnicity** Caucasian

**Morphology** Epithelial-like

**Growth properties** Adherent

### Regulatory Data

**Citation** Calu-6 (Cytion catalog number 300135)

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**Biosafety level** 1**NCBI\_TaxID** 9606**CellosaurusAccession** CVCL\_0236**Biomolecular Data****Protein expression** P53 negative**Isoenzymes** Me-2, 1, PGM3, 1, PGM1, 2, ES-D, 1, AK-1, 1, GLO-1, 2, G6PD, B, Phenotype Frequency Product: 0.0031**Tumorigenic** Yes, in nude mice. Forms poorly differentiated carcinoma**Mutational profile** Calu-6 cells carry a mutation in KRAS codon 61, c.181C>A p.(Gln61Lys). NRAS or BRAF mutation was not detected.**Karyotype** The stemline chromosome number is hypotriploid and the 2S component occurred at 5.8%. Modal chromosome number is 59. Fourteen marker chromosomes (constitutive) were common to most S metaphases. No Y chromosome was detected in the QM stained preparation.**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** 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**  $2 \times 10^4$  cells/cm<sup>2</sup> will result in a 90% confluent monolayer in about 4 days**Fluid renewal** 2 to 3 times per week

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### Post-Thaw Recovery

After thawing, plate the cells at  $5 \times 10^4$  cells/cm<sup>2</sup> and allow the cells to recover from the freezing process and to adhere for at least 48 hours.

### 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.