

## A549-RFP Cells | 305659

## General information

## Description

A549-RFP is a fluorescently labeled derivative of the human A549 lung adenocarcinoma cell line, engineered to constitutively express red fluorescent protein (RFP) for real-time visualization and tracking. The parental A549 line was established from a lung adenocarcinoma derived from an adult donor and exhibits epithelial morphology with adherent growth characteristics. A549 cells retain features of type II alveolar epithelial cells, including expression of cytokeratins and the capacity for surfactant-associated protein production. The introduction of a stable RFP expression cassette enables continuous fluorescence without significantly altering the intrinsic proliferative and metabolic properties of the parental line, making A549-RFP suitable for longitudinal imaging studies.

Functional characterization of A549 cells within large cancer cell panels has demonstrated that cell size, protein content, and protein synthesis rate are positively correlated with cell volume, and that larger cells tend to proliferate more slowly. In comparative analyses, A549 cells are positioned among relatively smaller, faster-proliferating epithelial cancer cell lines, in contrast to larger, more mesenchymal-like cells that display higher vimentin expression and lower E-cadherin levels. These metabolic and phenotypic distinctions are relevant for experimental interpretation, as protein synthesis rates and metabolic fluxes scale with cell size and influence sensitivity to agents targeting proliferation or mTOR-regulated anabolic pathways. The RFP modification preserves the suitability of A549 cells for such metabolic and pharmacologic investigations while enabling direct visualization.

A549-RFP is widely used in co-culture systems, orthotopic and ectopic xenograft models, and invasion or metastasis assays where fluorescent labeling facilitates discrimination of tumor cells from stromal or host components. The stable red fluorescence supports applications including live-cell imaging, high-content screening, flow cytometry-based quantification, and in vivo optical imaging. As a traceable variant of a well-characterized lung adenocarcinoma model, A549-RFP provides a robust platform for studying tumor cell proliferation, epithelial-mesenchymal transition, drug response, and tumor-microenvironment interactions in both in vitro and in vivo settings.

**Genetic modification:** Stably modified by replication-incompetent lentiviral transduction to express a red fluorescent protein (RFP) reporter; maintained as a polyclonal population under puromycin selection (1–5 µg/mL). S1/BSL-1 containment.

**Organism** Human

**Tissue** Lung

**Disease** Lung adenocarcinoma

**Synonyms** A 549, A549, NCI-A549, A549/ATCC, A549 ATCC, A549ATCC, hA549

## Characteristics

**Age** 58 years

**Gender** Male

**A549-RFP Cells | 305659****Ethnicity** Caucasian**Growth properties** Adherent**Regulatory Data****Citation** A549-RFP (Cytion catalog number 305659)**Biosafety level** 1**NCBI\_TaxID** 9606**CellosaurusAccession** CVCL\_0023**GMO Status** GMO-S1: This cell line contains a stably integrated red fluorescent protein (RFP) reporter introduced via replication-incompetent lentiviral transduction. The resulting polyclonal cell population was maintained under puromycin selection (1–5 µg/mL). S1 containment is required. This classification applies only within Germany and may differ elsewhere.**Biomolecular Data****Antigen expression** RFP (red fluorescent protein)**Mutational profile** Mutation: p.Gly12Ser, Homozygous; Mutation: p.Gln37Ter, Homozygous**Handling****Culture Medium** DMEM:Ham's F12 (1:1), w: 3.1 g/L Glucose, w: 2.5 mM L-Glutamine, w: 15 mM HEPES, w: 0.5 mM Sodium pyruvate, w: 1.2 g/L NaHCO<sub>3</sub> (Cytion article number 820400a)**Supplements** Supplement the medium with 10% FBS**Dissociation Reagent** Accutase**Doubling time** 20-40 hours**Freeze medium** As a cryopreservation medium, we use complete growth medium + 10% DMSO for adequate post-thaw viability.

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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  $-150^{\circ}\text{C}$  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  $37^{\circ}\text{C}$  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  $200 \times g$  for 5 minutes, carefully discard the supernatant containing freezing medium.
7. Follow the procedure described under Post-Thaw Recovery

### Incubation Atmosphere

$37^{\circ}\text{C}$ , 5%  $\text{CO}_2$ , humidified atmosphere.

### Shipping Conditions

Cryopreserved cell lines are shipped on dry ice in validated, insulated packaging with sufficient refrigerant to maintain approximately  $-78^{\circ}\text{C}$  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  $-196^{\circ}\text{C}$ . Storage at  $-80^{\circ}\text{C}$  is acceptable only as a short interim step before transfer to liquid nitrogen.

## Quality Control & Molecular Analysis