

A549/GFP Cell Line

CATALOG NUMBER: AKR-209

STORAGE: Liquid nitrogen

Note: For best results begin culture of cells immediately upon receipt. If this is not possible, store at -80°C until first culture. Store subsequent cultured cells long term in liquid nitrogen.

QUANTITY & CONCENTRATION: 1 mL, 1×10^6 cells/mL in 70% DMEM, 20% FBS, 10% DMSO

Background

The A549 cell line was first developed in 1972 through the removal and culturing of cancerous lung tissue in the explanted tumor of 58-year-old male. A549 cells are human alveolar basal epithelial cells. They are squamous in nature and responsible for the diffusion of substances across the alveoli of lungs. In vitro they grow adherently as a monolayer, and in vivo they induce tumors in athymic mice. A549 cells could synthesize lecithin with a high percentage of desaturated fatty acids utilizing the cytidine diphosphocholine pathway. Our A549/GFP cell line stably expresses GFP; the gene was introduced using lentivirus.

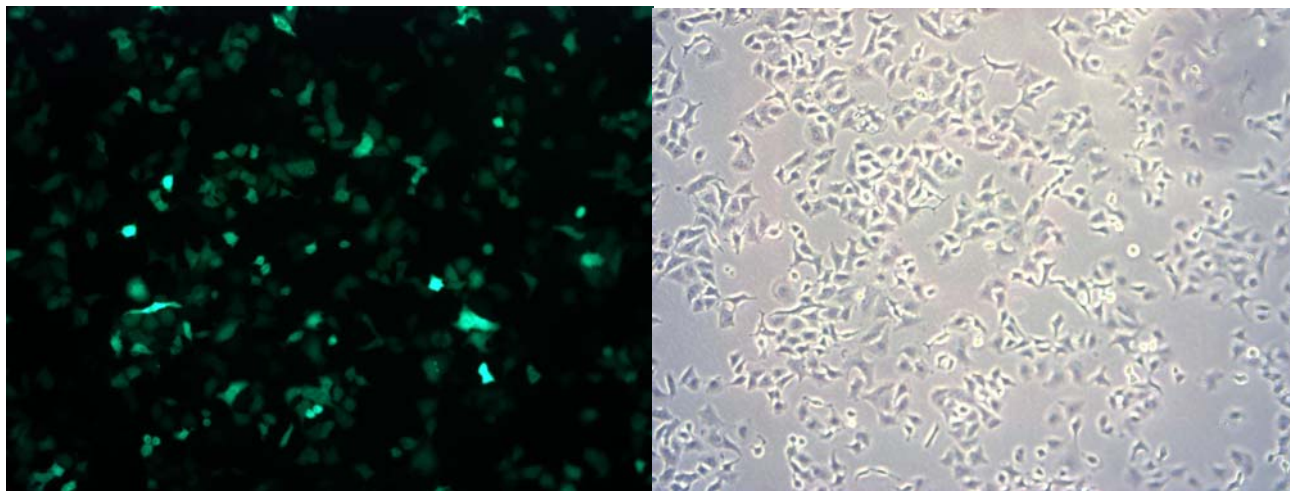


Figure 1. A549/GFP Cell Line. Left: GFP Fluorescence; Right: Phase Contrast.

Quality Control

This cryovial contains at least 1.0×10^6 A549/GFP cells as determined by morphology, trypan-blue dye exclusion, and viable cell count. The A549/GFP cells are tested free of microbial contamination.

Medium

1. Culture Medium: D-MEM (high glucose), 10% fetal bovine serum (FBS), 0.1 mM MEM Non-Essential Amino Acids (NEAA), 2 mM L-glutamine, 1% Pen-Strep.
2. Freeze Medium: 70% DMEM, 20% FBS, 10% DMSO.

Methods

Establishing A549/GFP Cultures from Frozen Cells

1. Place 10 mL of complete DMEM or McCoy's 5A growth medium in a 50-mL conical tube. Thaw the frozen cryovial of cells within 1–2 minutes by gentle agitation in a 37°C water bath. Decontaminate the cryovial by wiping the surface of the vial with 70% (v/v) ethanol.
2. Transfer the thawed cell suspension to the conical tube containing 10 ml of growth medium.
3. Collect the cells by centrifugation at 1000 rpm for 5 minutes at room temperature. Remove the growth medium by aspiration.
4. Resuspend the cells in the conical tube in 15 mL of fresh growth medium by gently pipetting up and down.
5. Transfer the 15 mL of cell suspension to a T-75 tissue culture flask. Place the cells in a 37°C incubator at 5% CO₂.
6. Monitor cell density daily. Cells should be passaged when the culture reaches 95% confluence.

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