

Datasheet for H1299/RFP Labeled Actin Cell Line

Catalog number: SL201

Product: H1299 cell line stably expressing RFP Labeled Actin.

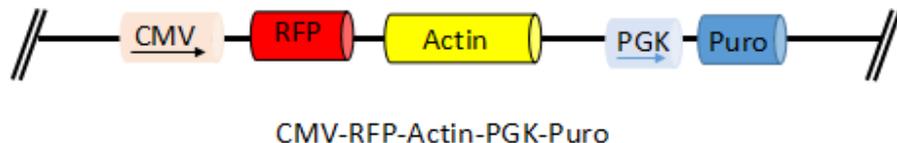
Description: This product is a cell line stably expressing the Actin. Actins are highly conserved proteins that are involved in various types of cell motility. This cell line also expresses RFP protein and the puromycin resistance gene.

Quantity: 1 vial of 2×10^6 cells; frozen

Shipping conditions: Dry ice

Storage conditions: Liquid nitrogen vapor phase. Remove the item from the dry ice packaging and check all items for damage and leakage. Place immediately into storage at or below -140°C , preferably into the liquid nitrogen vapor phase, until use.

Transgene integration:



Source of parental line:

H1299
Organism: *Homo sapiens*, human
Tissue: lung
Cell type: Epithelial

Quality control: >95% viability before freezing. All cells were tested and found to be free of mycoplasma, bacteria, viruses, and other toxins.

Safety instructions: To ensure safety, protective gloves, clothing, and a face mask should be worn when handling frozen vials. Some leakage may occur into the vial during storage. The liquid nitrogen will be converted to gas upon thawing. Due to the nature of nitrogen gas, pressure may build within the vial and possibly result in the vial exploding or losing its cap. This may cause flying debris.

Thawing procedure: The vial of cells should be thawed in a 37 °C water bath with gentle agitation. For optimal performance, the vial should be thawed in under two minutes. Ensure that the cap of the vial did not loosen upon thawing, and re-tighten as needed. Spray the vial with 70% EtOH and wipe off. Repeat. Using aseptic technique, add the contents of the vial to 9 ml of complete growth medium (without selection). Centrifuge for 5 min. at 125 x g. Aspirate the medium, being careful not to disturb the pellet. Resuspend in 10 mL of complete growth medium, and place into a culture vessel of your choice. Only add selection to the medium after 24 hours in culture.

Culture conditions:

Complete Growth Medium

The base medium for this cell line is DMEM. For optimal growth and maintenance of selection, add the following components to the base medium: dialyzed fetal bovine serum to a final concentration of 10%.

Selection

Puromycin to a final concentration of 2 µg/mL

Culture temperature:

37 °C with 5% CO₂

Subculture:

Replace culture medium with selection-free medium and incubate for up to 6 hours. Rinse the cells with PBS without cations, digest cells with 0.25% (w/v) Trypsin-EDTA (0.53 mM) solution and split at 1:3 to 1:10 ratio.

Cryopreservation: Freeze slowly in complete growth medium supplemented with 5% (v/v) DMSO.

Mycoplasma: Negative
(MycoGuard™ Mycoplasma Bioluminescent Detection Kit from GeneCopoeia)

Product QC:

1. Transduction

(1) Plate cells

Trypsinize and count the H1299 cells before one day. Adjust the cell concentration and seed H1299 cells in a 6-well plate, The number of cells plated in each well should be determined so that are 50-60% confluent at the time of transduction.

(2) Add Lentiviral particles

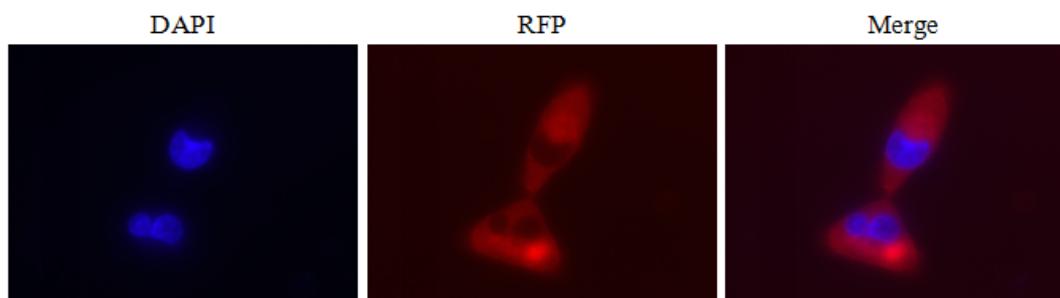
Add LP501 Lentiviral particles in H1299 cells according to the MOI=1. (LP501, Lentiviral particles of RFP-Actin, GeneCopoeia: 1.57×10^8 TU/ml)

2. Drug selection

Trypsinize the H1299 cells into 10cm² dish post-transduction 72 hours, add the puromycin in dish with final concentration 2 µg/mL. observe the drug selection after 3 days, Change the final concentration of puromycin (1 µg/mL) when the un-transduction cells were died.

3. Cells climbing to the carry sheet glass

Fluorescence: Image of H1299/RFP Labeled Actin Cell Line. Observing the cells climbing to the carry sheet glass, which are stained with DAPI.



Citation of product: If use of this item results in a publication, please use this information:
H1299/RFP Labeled Actin Cell Line (SL201, GeneCopoeia, Inc., Rockville, MD).



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