Certificate of Analysis



RetroPack™ PT67 Cell Line

Catalog No. Amount Lot Number

631510 1 ml Specified on product label.

Description

An NIH 3T3-derived cell line designed for the production of infectious, replication-incompetent virus. PT67 contains the Moloney murine leukemia virus (MoMuLV) gag, pol, and env (10A1-derived) genes. Transfection with a retroviral vector containing the retroviral packaging signal and a target gene allows production of replication-incompetent virus. (Cepko and Pear 1996; Coffin, Hughes, and Varmus 1997)

Package Contents

• 1 ml RetroPack PT67 Cell Line (2 x 10⁶ cells/tube)

Storage Conditions

• Store cells in liquid nitrogen (-196°C) or in a -150°C freezer

Shelf Life

• 1 year from date of receipt under proper storage conditions.

Storage Medium

• Cell Freezing Medium-DMSO 1x (Sigma-Aldrich Co., Cat. No. C6164)

Shipping Conditions

Dry ice

Product Documents

Documents for our products are available for download at <u>takarabio.com/manuals</u> The following documents apply to this product:

• Retroviral Gene Transfer and Expression User Manual

Cell Type Information

NIH 3T3-derived cell line stably expressing the retroviral gag and pol genes, cointroduced with the thymidine kinase gene; and the 10A1 envelope gene, cointroduced with the dihydrofolate reductase gene.

Recommended Cell Culture Medium

90% DMEM; 10% fetal bovine serum; 4 mM L-glutamine; 100 units/ml penicillin G sodium; 100 μ g/ml streptomycin sulfate; and 1 mM sodium pyruvate.

Takara Bio USA, Inc.

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RetroPackTM PT67 Cell Line

Recommendations for Thawing Frozen Cells

We recommend initiating the culture as soon as possible upon receipt. If the cells cannot be thawed and cultured immediately upon receipt, the vial should be held at temperatures below –79°C, preferably in liquid nitrogen vapor.

- 1. Thaw rapidly by placing tube of frozen culture in a 37°C water bath.
- 2. To reduce osmotic shock, dilute the cell suspension in 1 ml of complete growth medium and transfer it to a sterile 15 ml tube.
- 3. Add 5 ml of complete medium and mix.
- 4. Add an additional 5 ml of complete medium and mix gently.
- 5. Pellet the cells for 10 min at 125g. Discard supernatant.
- 6. Resuspend the pellet in 10 ml of complete growth medium and seed culture into a flask or culture dish.

References

Cepko, C. & Pear, W. Overview of the Retrovirus Transduction System. *Curr. Protoc. Mol. Biol.* **36,** 9.9.1-9.9.16 (1996). Coffin, J.M. et al (eds). *Retroviruses*. (Cold Spring Harbor Laboratory Press, 1997).

Quality Control Data

Functional Tests

The RetroPack PT67 cell line was functionally tested for its ability to produce infectious virus. RetroPack PT67 cells were transfected with pSIREN-RetroQ-ZsGreen RetroviralVector. After 48 hours, virus was collected and overlaid on HT1080 cells. After three days, the HT1080 cells were analyzed by flow cytometry to confirm retroviral infection. The titer of virus produced by RetroPack PT67 cells was at least 10⁵ cfu/ml.

Mycoplasma Contamination Test

This lot of the RetroPack PT67 Cell Line was tested and found to be free of *Mycoplasma* contamination.

NOTE: The viral supernatants produced by these retroviral systems could, depending on your cloned insert, contain potentially hazardous recombinant virus. Due caution must be exercised in the production and handling of recombinant retrovirus. Appropriate NIH, regional, and institutional guidelines apply. The User Manual contains other general information and precautions.

It is certified that this product meets the above specifications, as reviewed and approved by the Quality Department.

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5/26/2022