

Product Description

Product Name: Nalm6-Fluc-Puro
 Catalog Number: CL151
 Lot Number: IMP020

Species: Human (*Homo sapiens*)
 Tissues: Peripheral blood
 Cell type: Lymphoma
 Parental cells: Nalm6 (ATCC® CRL-3273™)*
 Morphology: Lymphocyte-like
 Growth mode: Suspension
 Reporter gene: Firefly luciferase (Fluc)
 Selection gene: Puromycin (Puro)

This cell line is derived from the human B cell precursor leukemia Nalm6 clone G5 cell line (ATCC® CRL-3273™). Parental Nalm6 cells were transduced with LV-Fluc-P2A-Puro (Imanis #LV012) encoding the firefly luciferase (Fluc) cDNA under the spleen focus-forming virus (SFFV) promoter and linked to the puromycin resistance gene (Puro) via a P2A cleavage peptide. High Fluc expressing cells were selected using puromycin followed by selection using a semi-solid methyl-cellulose-based medium. The lentiviral vector is a self-inactivating (SIN) vector in which the viral enhancer and promoter have been deleted. Transcription inactivation of the LTR in the SIN provirus increases biosafety by preventing mobilization by replication competent viruses and enables regulated expression of the genes from the internal promoters without *cis*-acting effects of the LTR¹.

* The ATCC trademark and any and all ATCC catalog numbers are trademarks of the American Type Culture Collection

Mycoplasma Testing

This cell line has been tested for mycoplasma contamination and is mycoplasma free.

Cell Line Authentication

Authentication of the parental Nalm6 cell line was confirmed by short tandem repeat (STR) profiling.

Recommended Uses

These cells are suitable for *in vitro* and *in vivo* experimentation.

The Fluc transgene facilitates *in vivo* noninvasive bioluminescent imaging of implanted cells.

References

¹Miyoshi et al. J Virol. 1998. 72:8150-8157.

Biosafety Notice

This cell line was generated by transduction with a lentiviral vector. Cell lines transduced with lentiviral vectors are classified as biosafety level 2 reagents and should be used under appropriate biosafety level for institutional guidelines.

Storage Instructions

Remove cells from the dry ice packaging and immediately store in the vapor phase above liquid nitrogen (below -130°C).

Complete Growth Medium

RPMI-1640 Medium (RPMI) containing 10 mM HEPES
 10% fetal bovine serum (FBS)
 1% Penicillin/Streptomycin
 1 µg/mL Puromycin

Puromycin should **NOT** be added to the medium until a culture has been well established from the thawed cells (about 1 week). It is also recommended that a backup frozen cell stock be generated (see below) before adding puromycin to the growth medium.

Caution! Typical commercial puromycin stocks are provided at a concentration of 10 mg/mL or 10,000X.

Thawing Instructions

1. Thaw cells by gently swirling in a 37°C water bath. To limit contamination, do not submerge the O-ring and cap.
2. When cells are ~70% thawed (~1 min), remove the vial and wipe down with 70% ethanol. Allow tube to dry completely.
3. In a biosafety cabinet, transfer the cells into a 15 mL conical tube containing 5 mL of complete growth medium. Centrifuge cells at ~300 x g for 3-5 min.
4. Remove supernatant and resuspend cells in complete growth medium to a final density of 1 x 10⁶ cells/mL. Transfer the cells to a T25 or T75 suspension culture flask.
5. Incubate the culture at 37°C with 5% CO₂.

Subculturing Instructions

Passage cells by dilution in fresh complete growth medium. If desired, use centrifugation to remove excess debris:

1. Pipet the cell suspension gently to dislodge any cells loosely attached to the culture flask. Transfer the desired volume (half, one fourth, etc.) of the cells to a conical tube.
2. Centrifuge at ~150 x g for 3 min. (Note: a short, low speed spin is recommended to limit the amount of cell debris in the pellet.)
3. Remove supernatant and resuspend cells in complete growth medium. Transfer to an appropriate sized flask.

The cells should be subcultured as needed to maintain a density between 5 x 10⁵ and 3 x 10⁶ cells/mL.

Freezing Medium

These cells can be amplified and used to generate additional frozen stocks. Cryopreservation of low passage frozen stocks is highly recommended. Frozen stocks should be preserved in a designated cryopreservation medium or in complete growth medium without puromycin supplemented with 5-10% DMSO.

Certificate of Analysis

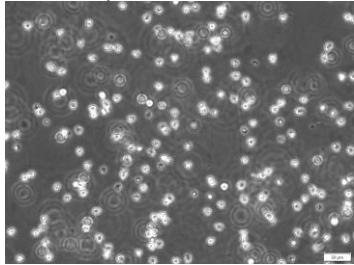
Testing performed by Imanis Life Sciences

Test description	Result
Post thaw viable cell recovery	94%
Cells per vial	$\sim 1 \times 10^7$
Sterility	No contamination detected
Mycoplasma	No contamination detected
Puromycin selection	Pass QC
Luciferase expression	Pass QC
Average doubling time	28.9 h*

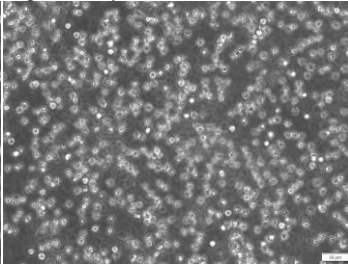
*Doubling time represents the average doubling time during logarithmic growth. This value should be used for general estimation only.

Morphology

Low density, 200X

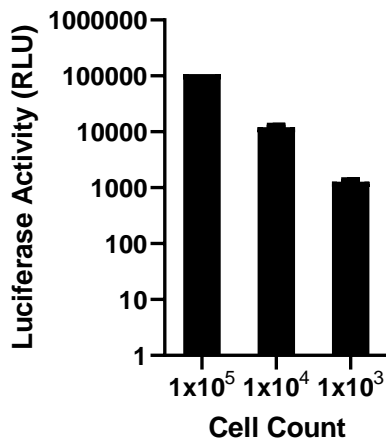


High density, 200X



Low- and high-density photos taken at various times after thawing.

Luciferase Expression



The indicated number of cells were placed in wells of a 96-well plate. After the addition of 15 mg/mL d-luciferin, bioluminescence was immediately read using a microplate reader.

Quality control by: AWD
 Quality Assurance by: RLV
 Effective Date: 18-May-2023

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