Raji-Fluc-EmGFP



Product Description

Product Name: Raji-Fluc-EmGFP

Catalog Number: CL159 Lot Number: CL-IM227

Species: Human (Homo sapiens)

Tissues: Lymphoblast Cell type: B Lymphoblast

Parental cells: Raji (ATCC® CCL-86™)*

Morphology: Lymphoblast Growth mode: Suspension

Reporter genes: Firefly luciferase (Fluc)

Emerald green fluorescent protein (EmGFP)

This is a cell line derived from the Raji cell line (ATCC® CCL-86TM). Parental Raji cells were transduced with LV-SFFV-Luc2-P2A-EmGFP (Imanis #LV050) encoding an enhanced firefly luciferase (Fluc) cDNA under the spleen focus-forming virus (SFFV) promoter and linked to the emerald green fluorescent protein (EmGFP) via a P2A cleavage peptide. High Fluc- and EmGFP- expresssing population was selected followed by selection with a methylcellulose-based semi-solid 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.

Mycoplasma Testing

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

Recommended Uses

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

The luciferase transgene facilitates *in vivo* noninvasive bioluminescent imaging of implanted cells. EmGFP is not recommended for whole animal in-live imaging. Rather, samples can be collected postmortem for analysis by conventional fluorescence microscopy or flow cytometry.

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) 10% Fetal Bovine Serum (FBS) 1% Penicillin/Streptomycin

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.
- 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 4-5 min.
- 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.

Note: These cells may exhibit an extended lag phase of growth. For optimal recovery use complete media with up to 20% FBS for thaw and initial passages.

Subculturing Instructions

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

- 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 the cells in complete growth medium. Transfer to an appropriately sized flask.

The cells should be subcultured as needed to maintain a density between 5×10^5 and 2×10^6 cells/mL.

Freezing Medium

These cells can be amplified and used to generate additional frozen stocks. Cryopreservation of low passage frozen stocks is recommended. Frozen stocks should be preserved in a designated cryopreservation medium or in complete growth medium supplemented with 5-10% DMSO.

Notes:

This cell line is positive for EBV.

These cells tend to form clumps, therefore daily gentle pipetting is recommended.

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

Raji-Fluc-EmGFP



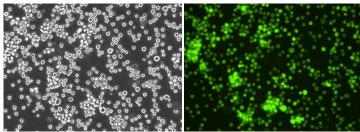
Certificate of Analysis

Testing performed by Imanis Life Sciences

Test description	Result
Post thaw viable cell recovery	86%
Viable cells per vial	~ 3.5 x 10 ⁶
Sterility	No contamination detected
Mycoplasma	No contamination detected
Luciferase expression	Pass QC
Fluorescence expression	Pass QC
Average doubling time	23 hours*

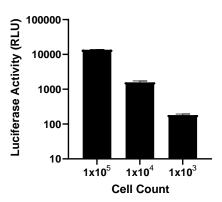
^{*}Doubling time represents the average doubling time during logarithmic growth. This value should be used for general estimation only.

Morphology



Cell photos taken at 200X magnification.

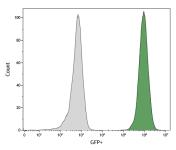
Luciferase Expression



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.

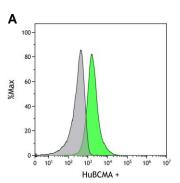
Quality control by: LAS/AWD **Quality Assurance by: RLV** Effective Date: 19-Sep-2023

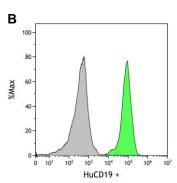
Fluorescence Expression

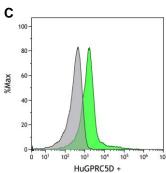


Raji-Fluc-EmGFP (green) or isotype control (Raji Parental; grey) cells were fixed with paraformaldehyde and analyzed by flow cytometry.

Expression Profiling of Surface Markers







Raji-Fluc-EmGFP cells were stained with anti-HuBCMA antibody (A), anti-HuCD19 antibody (B), or anti-HuGPRC5D antibody (C) and analyzed by flow cytometry; stained cells shown in green and isotype control shown in grey.

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