B16F10-Fluc-Neo/eGFP-Puro

Product Description

Product Name: B16F10-Fluc-Neo/eGFP-Puro
Catalog Number: CL068
Lot Number: CL-IM61

Shipping conditions: Dry ice
Storage conditions: Store in vapor phase above liquid nitrogen

Species: Mouse (*Mus musculus*)
Cell type: Melanoma
Morphology: Epithelial
Growth mode: Adherent
Reporter genes: Firefly luciferase (Fluc)
Enhanced green fluorescent protein (eGFP)
Selection genes: Neomycin (Neo)
Puromycin (Puro)
Media: DMEM, 10% FBS, 1% Penicillin/Streptomycin, 1 µg/mL puromycin, 0.8 mg/mL G418
Subculture: Split confluent culture 1:10 every 3-4 days using trypsin/EDTA
Incubation: 37°C with 5% CO_2

Description: B16F10-Fluc-Neo/eGFP-Puro is a polyclonal population of the murine melanoma cell line B16F10 transduced with lentiviral vectors encoding 1) the firefly luciferase (Fluc) cDNA under the spleen focus-forming virus (SFFV) promoter linked to the neomycin resistance gene (Neo) via a P2A cleavage peptide (Imanis #LV011) and 2) the enhanced green fluorescent protein (eGFP) cDNA under the SFFV promoter and the puromycin resistance gene under the phosphoglycerate kinase (PGK) promoter (Imanis #LV031). High Fluc and eGFP expressing cells were selected using G418 and puromycin. The lentiviral vectors are self-inactivating (SIN) vectors 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 LTR (Miyoshi et al., J Virol. 1998).

Cell line Authentication: Authentication of the parental B16F10 cell line was performed by Short tandem repeat (STR) profiling with 27 STR loci. STR profiling of B16F10 cells are verified and there is no interspecies cross contamination detected.

It has been estimated that ~18-36% of cell lines utilized in biomedical research are contaminated or completely misidentified (Hughes et al., BioTechniques 2007). Consequently, verification of cell line identity is of critical significance. Several funding organizations, including NIH, and major publishers, such as those affiliated with the American Association for Cancer Research (AACR), have established requirements for cell line authentication prior to publication. More information can be found in the links below.
https://grants.nih.gov/grants/guide/notice-files/NOT-OD-08-017.html
http://www.aacrjournals.org/site/InstrAuthors/ifora.xhtml#celllineuse

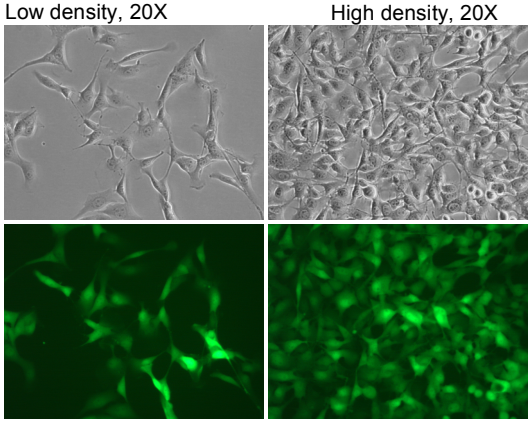
Certificate of Analysis

Testing performed by Imanis Life Sciences:

Test description	Result
Post thaw viable cell recovery	Pass QC
Sterility	No contamination detected
Mycoplasma	No contamination detected
Puromycin selection	Pass QC
Neomycin selection	Pass QC
Luciferase expression	Pass QC
Fluorescence expression	Pass QC

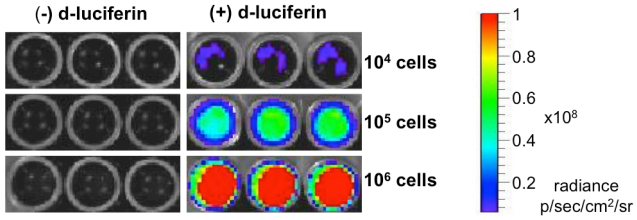
Morphology:

Low density, 20X High density, 20X



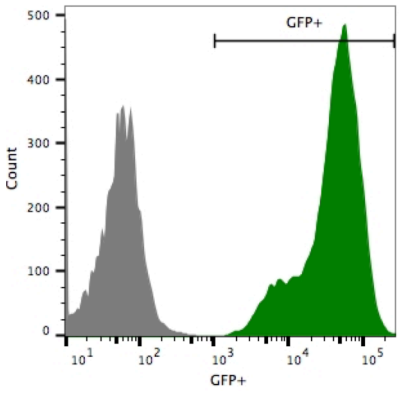
Low and high density photos taken 26 and 46 hours after thawing, respectively.

Luciferase Expression:



10^4, 10^5, or 10^6 cells were placed in wells of a 96-well plate and 0.3 mg of d-luciferin was added to the indicated wells. The plate was immediately imaged using a Xenogen IVIS Spectrum.

Fluorescence Expression:



B16F10-Fluc-Neo/eGFP-Puro (green) or isotype control (B16F10-Fluc-Neo/mNIS-Puro; grey) cells were fixed with paraformaldehyde and analyzed by flow cytometry (20,000 events).

Quality control by: RLV
Quality Assurance by: SPR
Effective Date: 11/6/15

Limited Product Warranty
This warranty limits our liability to replacement of this product. No other warranties of any kind, express or implied, including, without limitation, implied warranties of merchantability or fitness for a particular purpose, are provided by Imanis. Imanis shall have no liability for any direct, indirect, consequential, or incidental damages arising out of the use, the results of use, or the inability to use this product.

For *in vitro* use only. This certificate is a declaration of analysis at the time of manufacture.

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