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Produktinformation

- Forschungsprodukte & Biochemikalien
- Zellkultur & Verbrauchsmaterial
- Diagnostik & molekulare Diagnostik
- Laborgeräte & Service

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See the following pages for more information!

Lieferung & Zahlungsart

siehe unsere Liefer- und Versandbedingungen

Zuschläge

- Mindermengenzuschlag
- Trockeneiszuschlag
- Gefahrgutzuschlag
- Expressversand

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Description

eGFP/Firefly Luciferase BCMA Knockout MM.1S Cell Line is a MM.1S cell Line constitutively expressing firefly (*Photinus pyralis*) luciferase and the enhanced GFP (eGFP) cassette under the control of a CMV promoter, where BCMA (B-Cell Maturation Antigen, or CD269) has been genetically removed using CRISPR/Cas9 genome editing. This cell line was generated by using the eGFP/Firefly Luciferase Lentivirus (BPS Bioscience #79980).

This cell line has been validated by genome sequencing, flow cytometry and luciferase activity.

Background

B-Cell Maturation Antigen (BCMA), also known as CD269, is a cell surface receptor of the TNF receptor superfamily that recognizes B-Cell Activating Factor (BAFF). BCMA is preferentially expressed on mature B-lymphocytes and Multiple Myeloma (MM) cells. BCMA is a highly attractive target antigen for immunotherapy not only because of its restricted expression in nonmalignant tissue, but also due to its almost universal expression on MM cells. Pre-clinical studies using CAR (Chimeric Antigen Receptor) T-cells targeting BCMA have demonstrated anti-MM activity, and in 2017, the FDA granted BCMA CAR T-Cell immunotherapy the breakthrough designation in treating Multiple Myeloma.

MM.1S cells are multiple myeloma human B lymphoblasts. The parent cell line MM.1 was established from the peripheral blood of a multiple myeloma patient who had become resistant to steroid-based therapy. MM.1S cells are sensitive to dexamethasone.

Application

- Control in cell killing assays.
- Suitable as a B-cell target of CAR-T or CAR-NK cells during optimization of CAR design.

Materials Provided

Components	Format
2 vials of frozen cells	Each vial contains >1 x 10^6 cells in 1 ml of Cell Freezing Medium (BPS Bioscience #79796)

Parental Cell Line

MM.1S human B lymphoblasts, suspension

Mycoplasma Testing

The cell line has been screened to confirm the absence of Mycoplasma species.

Materials Required but Not Supplied

These materials are not supplied with the cell line but are necessary for cell culture and cellular assays. BPS Bioscience's reagents are validated and optimized for use with this cell line and are highly recommended for best results. Media components are provided in the Media Formulations section below.

Materials Required for Cell Line Culture

Name	Ordering Information
Thaw Medium 2	BPS Bioscience #60184
Growth Medium 2E	BPS Bioscience #79638

Materials Used in Cellular Assay

Name	Ordering Information
Thaw Medium 2	BPS Bioscience #60184
Growth Medium 2E	BPS Bioscience #79638
96-well Tissue Culture-treated White Clear-bottom Assay plate	Corning #3610
ONE-Step™ Luciferase Assay System	BPS Bioscience #60690
Luminometer	

Storage Conditions

Cells are shipped in dry ice and should immediately be thawed or stored in liquid nitrogen upon receipt. Do not use a -80°C freezer for long term storage. Contact technical support at support@bpsbioscience.com if the cells are not frozen in dry ice upon arrival.

Media Formulations

For best results, the use of validated and optimized media from BPS Bioscience is *highly recommended*. Other preparations or formulations of media may result in suboptimal performance.

Media Required for Cell Line Culture

Thaw Medium 2 (BPS Bioscience #60184):
RPMI 1640 (ATCC modification) medium supplemented with 10% FBS and 1% Penicillin/streptomycin.

Growth Medium 2E (BPS Bioscience #79638):
RPMI 1640 (ATCC modification) medium supplemented with 10% FBS, 1% Penicillin/streptomycin, plus 0.5 µg/ml of Puromycin Dihydrochloride.

Cell Culture Protocol

Cell Thawing

1. Retrieve a cell vial from liquid nitrogen storage. Keep on dry ice until ready to thaw.

2. When ready to thaw, swirl the vial of frozen cells for approximately 60 seconds in a 37°C water bath. Once cells are thawed (it may be slightly faster or slower than 60 seconds), quickly transfer the entire content of the vial to an empty 50 ml conical tube.

   Note: Leaving the cells in the water bath at 37°C for too long will result in rapid loss of viability.

3. Using a 10 ml serological pipette, slowly add 10 ml of pre-warmed Thaw Medium 2 to the conical tube containing the cells. Thaw Medium 2 should be added dropwise while gently rocking the conical tube to permit gentle mixing and avoid osmotic shock.

4. Immediately spin down the cells at 300 *x g* for 5 minutes, remove the medium and resuspend the cells in 5 ml of pre-warmed Thaw Medium 2.

5. Transfer the resuspended cells to a T25 flask or T75 flask and incubate at 37°C in a 5% CO_2 incubator.

6. After 24 hours of culture, check for viability. For a T25 flask, add 3-4 ml of fresh Thaw Medium 2 and continue growing culture in a 5% CO_2 incubator at 37°C until the cells are ready to passage.

7. Cells should be passaged before they reach 2 x 10^6 cells/ml. At first passage and subsequent passages, use Growth Medium 2E.

Cell Passage

Dilute the cell suspension into new culture vessels at no less than 0.2 x 10^6 cells/ml of Growth Medium 2E. The sub-cultivation ratio should be calculated so that cells are maintained between 0.2 x 10^6 cells/ml and 2 x 10^6 cells/ml.

Cell Freezing

1. Spin down the cells at 300 *x g* for 5 minutes, remove the medium and resuspend the cell pellet in 4°C Cell Freezing Medium (BPS Bioscience #79796) at a density of ~2 x 10^6 cells/ml.

2. Dispense 1 ml of cell suspension into each cryogenic vial. Place the vials in an insulated container for slow cooling and store at -80°C overnight.

3. Transfer the vials to liquid nitrogen the next day for long term storage.

Note: It is recommended to expand the cells and freeze at least 10 vials at an early passage for future use.

Validation Data

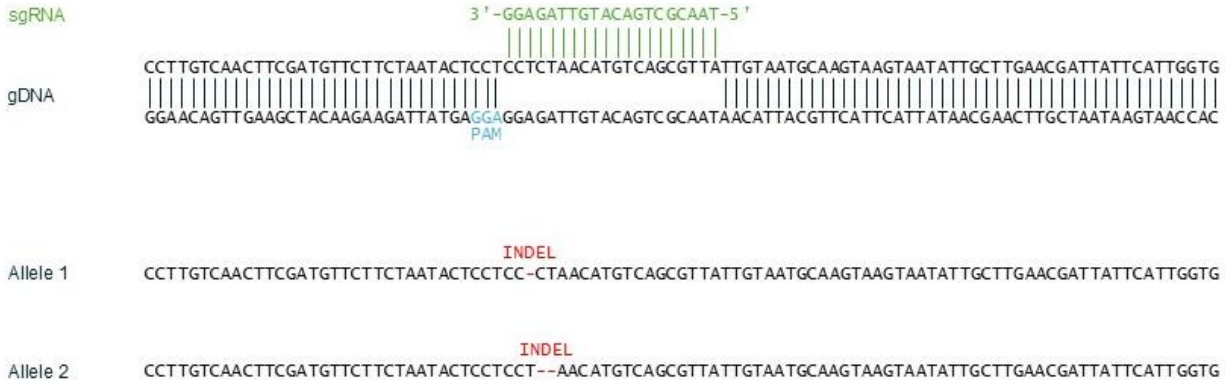

Figure 1. Genomic sequencing of BCMA in the eGFP/Firefly Luciferase BCMA Knockout MM.1S Cell Line.
Genomic DNA from the eGFP/Firefly Luciferase BCMA Knockout MM.1S cells was isolated and sequenced. The PAM (Protospacer Adjacent Motif) is shown in blue, the sgRNA (synthetic guide RNA) is shown in green, and the Indels (Insertions/Deletions) in the two BCMA alleles are highlighted in red. The BCMA genomic DNA is labeled as gDNA.

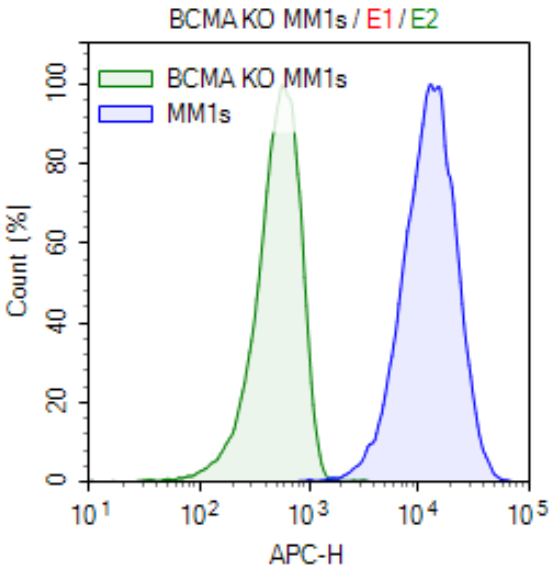

Figure 2. Flow cytometry analysis of BCMA expression in eGFP/Firefly Luciferase BCMA Knockout MM.1S Cell Line.
Cells were stained with APC anti-human CD269 (BCMA) Antibody (BioLegend #357505) and analyzed by flow cytometry. The parental MM.1S cells are shown in blue, and the eGFP/Firefly Luciferase BCMA Knockout MM.1S cells are shown in green. The y axis shows the % of cells, while the x axis represents the fluorophore intensity.

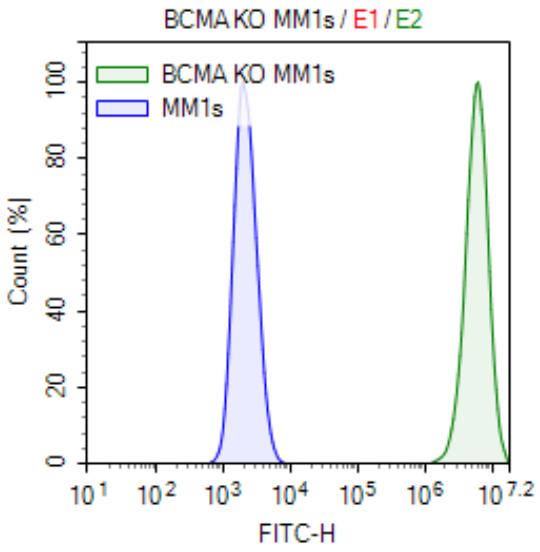

Figure 3. Flow cytometry analysis of GFP expression in eGFP/Firefly Luciferase BCMA Knockout MM.1S Cell Line.
Cells were analyzed by flow cytometry for GFP (FITC) expression. The parental MM.1S cells are shown in blue, and the eGFP/Firefly Luciferase BCMA Knockout MM.1S cells are shown in green. The y axis shows the % of cells, while the x axis represents the fluorophore intensity.

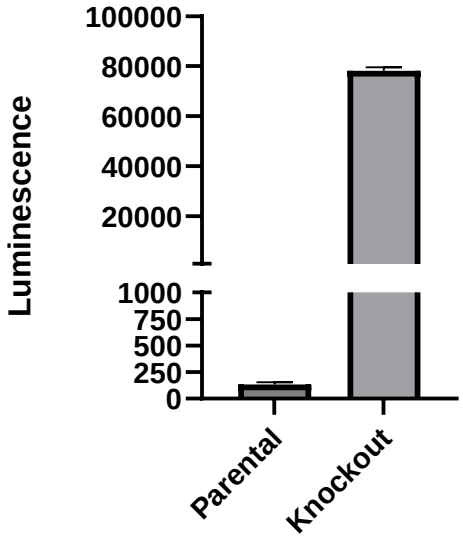

Figure 4. Luciferase activity in eGFP/Firefly Luciferase BCMA Knockout MM.1S Cell Line.
Parental MM.1S cells and eGFP/Firefly Luciferase BCMA Knockout MM.1S cells were seeded into a 96-well plate at 5,000 cells/well in 50 µl of Thaw Medium 2. Luciferase activity was measured using the ONEStep™ Luciferase Assay System.

Data shown is representative. For lot-specific information, please contact BPS Bioscience, Inc. at support@bpsbioscience.com.

References

Greenstein S., 2003 *et al*. *Exp. Hematol.* 31: 271-282.

License Disclosure

Visit bpsbioscience.com/license for the label license and other key information about this product.

Troubleshooting Guide

Visit bpsbioscience.com/cell-line-faq for detailed troubleshooting instructions. For all further questions, please email support@bpsbioscience.com.

Related Products

Products	*Catalog #*	*Size*
eGFP/Firefly Luciferase MM.1S Cell Line	78376	2 vials
BCMA CHO Recombinant Cell Line (High or Low Expression)	79500	2 vials
BCMA/ Luciferase – CHO Recombinant Cell Line	79724	2 vials
Anti-BCMA CAR-T Cells	78660	1 vial/ 5 vials
Dual Epitope Anti-BCMA CAR-T Cells	78790	1 vial/ 5 vials
BCMA Lentivirus	78714	500 µl x 2

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