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Produktinformation

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

Weitere Information auf den folgenden Seiten!
See the following pages for more information!

Lieferung & Zahlungsart

siehe unsere Liefer- und Versandbedingungen

Zuschläge

- Mindermengenzuschlag
- Trockeneiszuschlag
- Gefahrgutzuschlag
- Expressversand

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Description

B2M Knockout A549 Cell Line is an A549 cell line where B2M (Beta-2-Microglobulin) has been genetically removed by CRISPR/Cas9 genome editing.

Background

Beta-2-Microglobulin is a required component of Major Histocompatibility Complex (MHC) class 1 molecules, which present peptide fragments from within the cell to cytotoxic T cells as part of the adaptive immune system. The protein forms amyloid fibrils in some pathological conditions. A mutation in this gene drives hypercatabolic hypoproteinemia. B2M plays an essential role both in governing MHC class I molecule stability and in promoting antigen binding and presenting the antigen to CD3/TCR complex of $CD8^+$ T cells.

Application

- Study the impact of B2M knock-down.
- Study T cell activation, antigen presentation, and immune responses.

Materials Provided

Components	Format
2 vials of frozen cells	Each vial contains >1 x 106 cells in 1 ml of Cell Freezing Medium (BPS Bioscience #79796)

Parental Cell Line

A549 is a human lung alveolar vessel carcinoma cell line. Adherent epithelial cells.

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.

Media Required for Cell Culture

Name	Ordering Information
Thaw Medium 6	BPS Bioscience #60183

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.

Note: Thaw Media do *not* contain selective antibiotics.

Cells should be grown at 37℃ with 5% CO_2. BPS Bioscience's cell lines are stable for at least 10 passages when grown under proper conditions.

Media Required for Cell Culture

Thaw Medium 6 (BPS Bioscience #60183):
DMEM medium supplemented with 10% FBS, 1% Penicillin/Streptomycin.

Cell Thawing

1. Swirl the vial of frozen cells for approximately 60 seconds in a 37°C water bath. As soon as the cells are thawed (it may be slightly faster or slower than 60 seconds), quickly transfer the entire contents of the vial to a tube containing 10 ml of pre-warmed Thaw Medium 6.

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

2. 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 6.

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

4. After 24 hours of culture, check for cell attachment and viability. Change medium to fresh Thaw Medium 6 and continue growing in a 5% CO_2 incubator at 37°C until the cells are ready to passage.

5. Replace media every 2-3 days until cells reach 90% confluency. At first passage and subsequent passages, use Thaw Medium 6.

Cell Passage

1. Aspirate the medium, wash the cells with phosphate buffered saline (PBS) without Ca^{2+}/Mg^{2+}, and detach the cells from the culture vessel with 0.25% Trypsin/EDTA.

2. Once the cells have detached, add Thaw Medium 6 and transfer to a tube.

3. Spin down cells at 300 *x g* for 5 minutes, remove the medium and resuspend the cells in Thaw Medium 6.

4. Seed into new culture vessels at the recommended sub-cultivation ratio of 1:2 to 1:10 once or twice per week.

Cell Freezing

1. Aspirate the medium, wash the cells with PBS without Ca^{2+}/Mg^{2+}, and detach the cells from the culture vessel with 0.25% Trypsin/EDTA.

2. Once the cells have detached, add Thaw Medium 6 and count the cells.

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

4. 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.

5. 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

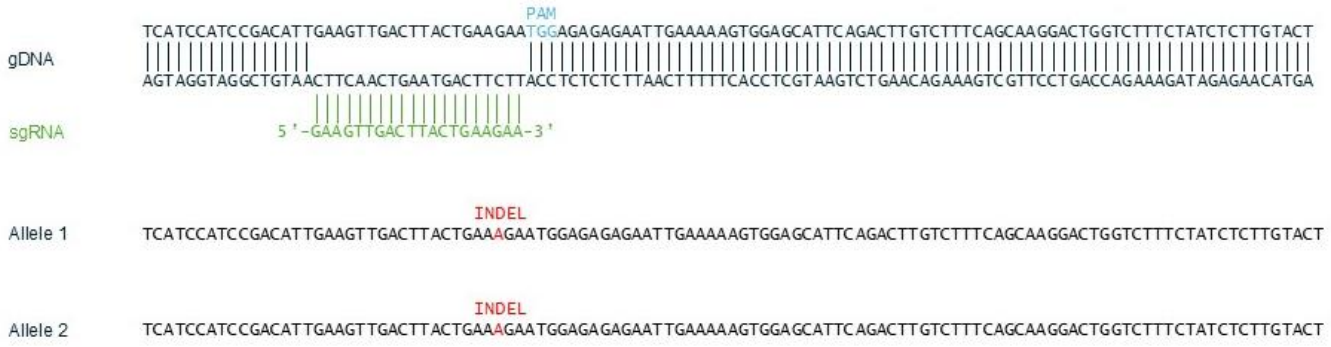

Figure 1: Genomic sequencing of B2M in the B2M Knockout A549 Cell Line.
Genomic DNA from the B2M Knockout A549 cells was isolated and sequenced. The PAM (Protospacer Adjacent Motif) is shown in blue, the sgRNA (synthetic guide RNA) is shown in green, and the Indels (Insertions/Deletions) in the two B2M alleles are highlighted in red. The B2M genomic DNA is labeled as gDNA.

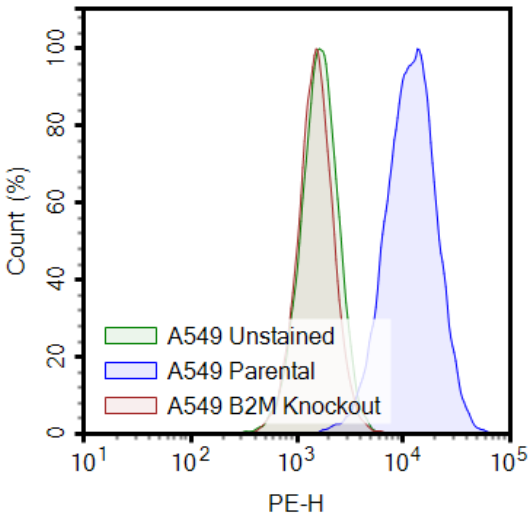

Figure 2. Expression of B2M in the B2M Knockout A549 cells.
B2M Knockout A549 cells (red) or parental A549 cells (blue) were stained with PE anti-human β2-microglobulin Antibody (BioLegend #395703) and analyzed by flow cytometry. Unstained parental A549 cells are shown in green. Y-axis is the % cell number. X-axis is the intensity of PE.

Data shown is representative.

License Disclosure

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

Troubleshooting

For lot-specific information and all other questions, please email visit https://bpsbioscience.com/contact. Visit bpsbioscience.com/cell-line-faq for detailed troubleshooting instructions.

Notes

The CRISPR/CAS9 technology is covered under numerous patents, including U.S. Patent Nos. 8,697,359 and 8,771,945, as well as corresponding foreign patents applications, and patent rights.

Related Products

Products	*Catalog #*	*Size*
B2M Knockout Jurkat Cell Line	82872	2 vials
B2M Knockout THP-1 Cell Line	78389	2 vials
B2M Knockout iPS Cell Line	82161	1 vial
B2M Knockout NFAT Luciferase Reporter Jurkat Cell Line	78363	2 vials
B2M (Human) CRISPR/Cas9 Lentivirus (Integrating)	78340	500 µl x 2
B2M (Human) CRISPR/Cas9 Lentivirus (Non-Integrating)	78341	500 µl x 2

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